

Figure 1

GATCAAACCTCTTTCCATTCAAGAGCCTCTGATTCAAGATTTAATGTTAACATTTGGAAGACAGTATTCAAGAAAAAAATTTCCTTAATAAAAATACAACTCAGATCCTCAAATATGAAACTGGTTGGGAACTCCATTTCATATTATTTCTTCTTGTTTC TTGCTACGTATAATTATTAATATCCTGACTAGGTTGTTGGAGGGTTATTACTTTCACTTACCATGCAGTCAAATCTAAAC TGCTTCACTGATGGTTACAGCATTCTGAGATAAGAATGGTACATCTAGAGAACATTGCCAAGGCCTAAGCACAGCAAAGGAA AATAAACACAGAATATAAAATGAGATAATCTAGCTAAACTATAACTCCCTTTAGAACTCCCAACCACATTGGATC

FIG. 2A

9	18	27	36	45	54													
5'	CAG AGA GGC TGT ATT TCA GTG CAG CCT GCC AGA CCT CTT CTG GAG GAA GAC TGG																	
	-----	-----	-----	-----	-----													
	63	72	81	90	99	108												
	ACA AAG GGG GTC ACA CAT TCC TTC CAT ACG GTT GAG CCT CTA CCT GCC TGG TGC																	
	-----	-----	-----	-----	-----	-----												
117	126	135	144	153	162													
TGG TCA CAG TTC AGC TTC TTC	<u>ATG</u>	ATG GTG GAT CCC AAT GGC AAT GAA TCC AGT																
	-----	-----	-----	-----	-----													
	M	M	V	D	P	N	G	N	E	S	S							
171	180	189	198	207	216													
GCT ACA TAC TTC ATC CTA ATA GGC CTC CCT GGT TTA GAA GAG GCT CAG TTC TGG																		
	-----	-----	-----	-----	-----													
	A	T	Y	F	I	L	I	G	L	P	G	L	E	E	A	Q	F	W
225	234	243	252	261	270													
TTG GCC TTC CCA TTG TGC TCC CTC TAC CTT ATT GCT GTG CTA GGT AAC TTG ACA																		
	-----	-----	-----	-----	-----													
	L	A	F	P	L	C	S	L	Y	L	I	A	V	L	G	N	L	T
279	288	297	306	315	324													
ATC ATC TAC ATT GTG CGG ACT GAG CAC AGC CTG CAT GAG CCC ATG TAT ATA TTT																		
	-----	-----	-----	-----	-----													
	I	I	Y	I	V	R	T	E	H	S	L	H	E	P	M	Y	I	F
333	342	351	360	369	378													
CTT TGC ATG CTT TCA GGC ATT GAC ATC CTC ATC TCC ACC TCA TCC ATG CCC AAA																		
	-----	-----	-----	-----	-----													
	L	C	M	L	S	G	I	D	I	L	I	S	T	S	S	M	P	K
387	396	405	414	423	432													
ATG CTG GCC ATC TTC TGG TTC AAT TCC ACT ACC ATC CAG TTT GAT GCT TGT CTG																		
	-----	-----	-----	-----	-----													
	M	L	A	I	F	W	F	N	S	T	T	I	Q	F	D	A	C	L
441	450	459	468	477	486													
CTA CAG ATT TTT GCC ATC CAC TCC TTA TCT GGC ATG GAA TCC ACA GTG CTG CTG																		
	-----	-----	-----	-----	-----													
	L	Q	I	F	A	I	H	S	L	S	G	M	E	S	T	V	L	L
495	504	513	522	531	540													
GCC ATG GCT TTT GAC CGC TAT GTG GCC ATC TGT CAC CCA CTG CGC CAT GCC ACA																		
	-----	-----	-----	-----	-----													
	A	M	A	F	D	R	Y	V	A	I	C	H	P	L	R	H	A	T
549	558	567	576	585	594													
GTA CTT ACG TTG CCT CGT GTC ACC AAA ATT GGT GTG GCT GCT GTG GTG CGG GGG																		
	-----	-----	-----	-----	-----													
	V	L	T	L	P	R	V	T	K	I	G	V	A	A	V	V	R	G
603	612	621	630	639	648													
GCT GCA CTG ATG GCA CCC CTT CCT GTC TTC ATC AAG CAG CTG CCC TTC TGC CGC																		
	-----	-----	-----	-----	-----													
	A	A	L	M	A	P	L	P	V	F	I	K	Q	L	P	F	C	R

FIG. 2B

657	666	675	684	693	702
TCC AAT ATC CTT TCC CAT TCC TAC TGC CTA CAC CAA GAT GTC ATG AAG CTG GCC					
S N I L S H S Y C L H Q D V M K L A					
711	720	729	738	747	756
TGT GAT GAT ATC CGG GTC AAT GTC GTC TAT GGC CTT ATC GTC ATC ATC TCC GCC					
C D D I R V N V V Y G L I V I I S A					
765	774	783	792	801	810
ATT GGC CTG GAC TCA CTT CTC ATC TCC TTC TCA TAT CTG CTT ATT CTT AAG ACT					
I G L D S L L I S F S Y L L I L K T					
819	828	837	846	855	864
GTG TTG GGC TTG ACA CGT GAA GCC CAG GCC AAG GCA TTT GGC ACT TGC GTC TCT					
V L G L T R E A Q A K A F G T C V S					
873	882	891	900	909	918
CAT GTG TGT GCT GTG TTC ATA TTC TAT GTA CCT TTC ATT GGA TTG TCC ATG GTG					
H V C A V F I F Y V P F I G L S M V					
927	936	945	954	963	972
CAT CGC TTT AGC AAG CGG CGT GAC TCT CCG CTG CCC GTC ATC TTG GCC AAT ATC					
H R F S K R R D S P L P V I L A N I					
981	990	999	1008	1017	1026
TAT CTG CTG GTT CCT CCT GTG CTC AAC CCA ATT GTC TAT GGA GTG AAG ACA AAG					
Y L L V P P V L N P I V Y G V K T K					
1035	1044	1053	1062	1071	1080
GAG ATT CGA CAG CGC ATC CTT CGA CTT TTC CAT GTG GCC ACA CAC GCT TCA GAG					
E I R Q R I L R L F H V A T H A S E					
1089	1098	1107	1116	1125	1134
CCC TAG GTG TCA GTG ATC AAA CTT CTT TTC CAT TCA GAG TCC TCT GAT TCA GAT					
P *					
1143	1152	1161	1170	1179	1188
TTT AAT GTT AAC ATT TTG GAA GAC AGT ATT CAG AAA AAA AAT TTC CTT AAT AAA					
1197	1206	1215	1224	1233	1242
AAA TAC AAC TCA GAT CCT TCA AAT ATG AAA CTG GTT GGG GAA TCT CCA TTT TTT					
1251	1260	1269	1278	1287	1296
CAA TAT TAT TTT CTT CTT TGT TTT CTT GCT ACA TAT AAT TAT TAA TAC CCT GAC					
1305	1314	1323	1332	1341	1350
TAG GTT GTG GTT GGA GGG TTA TTA CTT TTC ATT TTA CCA TGC AGT CCA AAT CTA					

FIG. 2C

1359	1368	1377	1386	1395	1404
AAC TGC TTC TAC TGA TGG TTT ACA GCA TTC TGA GAT AAG AAT GGT ACA TCT AGA					
-----	-----	-----	-----	-----	-----
1413	1422	1431	1440	1449	1458
GAA CAT TTG CCA AAG GCC TAA GCA CGG CAA AGG AAA ATA AAC ACA GAA TAT AAT					
-----	-----	-----	-----	-----	-----
1467	1476	1485	1494	1503	1512
AAA ATG AGA TAA TCT AGC TTA AAA CTA TAA CCT CCT CTT CAG AAC TCC CAA CCA					
-----	-----	-----	-----	-----	-----
1521	1530	1539	1548	1557	1566
CAT TGG ATC TCA GAA AAA TGC TGT CTT CAA AAT GAC TTC TAC AGA GAA GAA ATA					
-----	-----	-----	-----	-----	-----
1575	1584	1593	1602	1611	1620
ATT TTT CCT CTG GAC ACT AGC ACT TAA GGG GAA GAT TGG AAG TAA AGC CTT GAA					
-----	-----	-----	-----	-----	-----
1629	1638	1647	1656	1665	1674
AAG AGT ACA TTT ACC TAC GTT AAT GAA AGT TGA CAC ACT GTT CTG AGA GTT TTC					
-----	-----	-----	-----	-----	-----
1683	1692	1701	1710	1719	1728
ACA GCA TAT GGA CCC TGT TTT TCC TAT TTA ATT TTC TTA TCA ACC CTT TAA TTA					
-----	-----	-----	-----	-----	-----
1737	1746	1755	1764	1773	1782
GGC AAA GAT ATT ATT AGT ACC CTC ATT GTA GCC ATG GGA AAA TTG ATG TTC AGT					
-----	-----	-----	-----	-----	-----
1791	1800	1809	1818	1827	1836
GGG GAT CAG TGA ATT AAA TGG GGT CAT ACA AGT ATA AAA ATT AAA AAA AAA AAA					
-----	-----	-----	-----	-----	-----
1845	1854	1863	1872	1881	1890
GAC TTC ATG CCC AAT CTC ATA TGA TGT GGA AGA ACT GTT AGA GAG ACC AAC AGG					
-----	-----	-----	-----	-----	-----
1899	1908	1917	1926	1935	1944
GTA GTG GGT TAG AGA TTT CCA GAG TCT TAC ATT TTC TAG AGG AGG TAT TTA ATT					
-----	-----	-----	-----	-----	-----
1953	1962	1971	1980	1989	1998
TCT TCT CAC TCA TCC AGT GTT GTA TTT AGG AAT TTC CTG GCA ACA GAA CTC ATG					
-----	-----	-----	-----	-----	-----
2007	2016	2025	2034	2043	2052
GCT TTA ATC CCA CTA GCT ATT GCT TAT TGT CCT GGT CCA ATT GCC AAT TAC CTG					
-----	-----	-----	-----	-----	-----
2061	2070	2079	2088	2097	2106
TGT CTT GGA AGA AGT GAT TTC TAG GTT CAC CAT TAT GGA AGA TTC TTA TTC AGA					
-----	-----	-----	-----	-----	-----
2115	2124	2133	2142	2151	2160
AAG TCT GCA TAG GGC TTA TAG CAA GTT ATT TAT TTT TAA AAG TTC CAT AGG TGA					
-----	-----	-----	-----	-----	-----
2169	2178	2187	2196	2205	2214
TTC TGA TAG GCA GTG AGG TTA GGG AGC CAC CAG TTA TGA TGG GAA GTA TGG AAT					
-----	-----	-----	-----	-----	-----
2223	2232	2241	2250	2259	2268
GGC AGG TCT TGA AGA TAA CAT TGG CCT TTT GAG TGT GAC TCG TAG CTG GAA AGT					
-----	-----	-----	-----	-----	-----
2277	2286	2295	2304	2313	2322
GAG GGA ATC TTC AGG ACC ATG CTT TAT TTG GGG CTT TGT GCA GTA TGG AAC AGG					
-----	-----	-----	-----	-----	-----
2331	2340	2349	2358	2367	2376
GAC TTT GAG ACC AGG AAA GCA ATC TGA CTT AGG CAT GGG AAT CAG GCA TTT TTG					
-----	-----	-----	-----	-----	-----

FIG. 2D

2385	2394	2403	2412	2421	2430												
CTT	CTG	AGG	GGC	TAT	TAC	CAA	GGG	TTA	ATA	GGT	TTC	ATC	TTC	AAC	AGG	ATA	TGA
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2439	2448	2457	2466	2475	2484												
CAA	CAG	TGT	TAA	CCA	AGA	AAC	TCA	AAT	TAC	AAA	TAC	TAA	AAC	ATG	TGA	TCA	TAT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2493	2502	2511	2520	2529	2538												
ATG	TGG	TAA	GTT	TCA	TTT	TCT	TTT	TCA	ATC	CTC	AGG	TTC	CCT	GAT	ATG	GAT	TCC
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2547	2556	2565	2574	2583	2592												
TAT	AAC	ATG	CTT	TCA	TCC	CCT	TTT	GTA	ATG	GAT	ATC	ATA	TTT	GGA	AAT	GCC	TAT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2601	2610	2619	2628	2637	2646												
TTA	ATA	CTT	GTA	TTT	GCT	GCT	GGA	CTG	TAA	GCC	CAT	GAG	GGC	ACT	GTT	TAT	TAT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2655	2664	2673	2682	2691	2700												
TGA	ATG	TCA	TCT	CTG	TTC	ATC	ATT	GAC	TGC	TCT	TTG	CTC	ATC	ATT	GAA	TCC	CCC
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2709	2718	2727	2736	2745	2754												
AGC	AAA	GTG	CCT	AGA	ACA	TAA	TAG	TGC	TTA	TGC	TTG	ACA	CCG	GTT	ATT	TTT	CAT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2763	2772	2781	2790	2799	2808												
CAA	ACC	TGA	TTC	CTT	CTG	TCC	TGA	ACA	CAT	AGC	CAG	GCA	ATT	TTC	CAG	CCT	TCT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2817	2826	2835	2844	2853	2862												
TTG	AGT	TGG	GTA	TTA	TTA	AAT	TCT	GGC	CAT	TAC	TTC	CAA	TGT	GAG	TGG	AAG	TGA
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2871	2880	2889	2898	2907	2916												
CAT	GTG	CAA	TTT	CTA	TAC	CTG	GCT	CAT	AAA	ACC	CTC	CCA	TGT	GCA	GCC	TTT	CAT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2925	2934	2943	2952	2961	2970												
GTT	GAC	ATT	AAA	TGT	GAC	TTG	GGA	AGC	TAT	GTG	TTA	CAC	AGA	GTA	AAT	CAC	CAG
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
2979	2988	2997	3006	3015	3024												
AAG	CCT	GGA	TTT	CTG	AAA	AAA	CTG	TGC	AGA	GCC	AAA	CCT	CTG	TCA	TTT	GCA	ACT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
3033	3042	3051	3060	3069	3078												
CCC	ACT	TGT	ATT	TGT	ACG	AGG	CAG	TTG	GAT	AAG	TGA	AAA	ATA	AAG	TAC	TAT	TGT
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----
3087	3096	3105	3114	3123	3132												
GTC	AAG	AAA															
-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----	-----

AAA A 3'

Figure 3: Protein Sequence for 101P3A11.

MVDPNGNESSATYFILIGLPGLEEAQFWLAFLPLCSLYLIAVLGNLTIIYIVRTEHSLHEPMYIFLCMLSGIDILI
STSSMPKMLAIFWFNSTTIQFDACLLQIFAIHSLSGMESTVLLAMAFDRYVAICHPLRHATVLTLPRTKIGV
AAVVRGAALMAPLPVFIKQLPFCRSNILSHSYCLHQDVMKLACDDIRVNVVYGLIVISAIGLDSLLISFSYL
LILKTVLGLTREAQAKAFGTCVSHVCASFIFYVPFIGLSMVHRFSKRRDSPLPVILANTYLLVPPVLPNPIVYG
VKTKEIRQRILRLFHVATHASEP

Figure 4**Alignment of 101P3A11 (Sbjct) with mouse olfactory receptor S25 (Query)**

Query: 34 GNYTVVTEFILLGLTDDITVSILFVVMFLIVYSVTLMGNLNIIVLIRTSPQLHTPMYLFL 93
GN + T FIL+GL L +Y + ++GNL II ++RT LH PMY+FL

Sbjct: 6 GNESSATYFILIGLPGLEEAQFWLAFLPLCSLYLIAVLGNLTIIYIVRTEHSLHEPMYIFL 65

Query: 94 SHLAFLDIGYSSSVTPIMLRGFLRKGTIFPVAGCVAQLCIVVAFGTSESFLLASMAYDRY 153
L+ +DI S+S P ML F T I C+ Q+ + + ES +L +MA+DRY

Sbjct: 66 CMLSGIDILISTSSMPKMLAIFWFNSTTIQFDACLLQIFAIHSLSGMESTVLLAMAFDRY 125

Query: 154 VAICSPLLYSTQMSSTVCILLVGTSYLGGVNAWIFTGCSLNLSFCGPNKINHFFCDYSP 213
VAIC PL ++T ++ + + + G L FC N ++H +C +

Sbjct: 126 VAICHPLRHATVLTLPRTVKIGVAAVVRGAALMAPLPVFIKQLPFCRSNILSHSYCLHQD 185

Query: 214 LLKLSCSHDFSFEVIPAISGSIIIVVTVFIIALSYVYILVSILKMRSTEGRQKAFSTCTS 273
++KL+C V I S I + +I+ SY+ IL ++L + + E + KAF TC S

Sbjct: 186 VMKLACDDIRVNYYGLIVIISAIGLDSLLISFSYLLILKTVLGL-TREAQAKAFGTCVS 244

Query: 274 HLTAVTLFFGTITFIYVMPQSSYSTDQNK---VVSVFYTVVIPMLNPLIYSFRNKEVKE 329
H+ AV +F+ + FI + +S ++ + + + Y +V P+LNP++Y + KE+++

Sbjct: 245 HVCAVFIFY--VPFIGLSMVHRFSKRRDSPLPVILANIYLLVPPVLNPIVYGVKTKEIRQ 302

Query: 330 AMKKL 334

+ +L

Sbjct: 303 RILRL 307

Figure 5:
101P3A11 Hydrophilicity profile
(Hopp T.P., Woods K.R., 1981. Proc. Natl. Acad. Sci. U.S.A. 78:3824-3828)

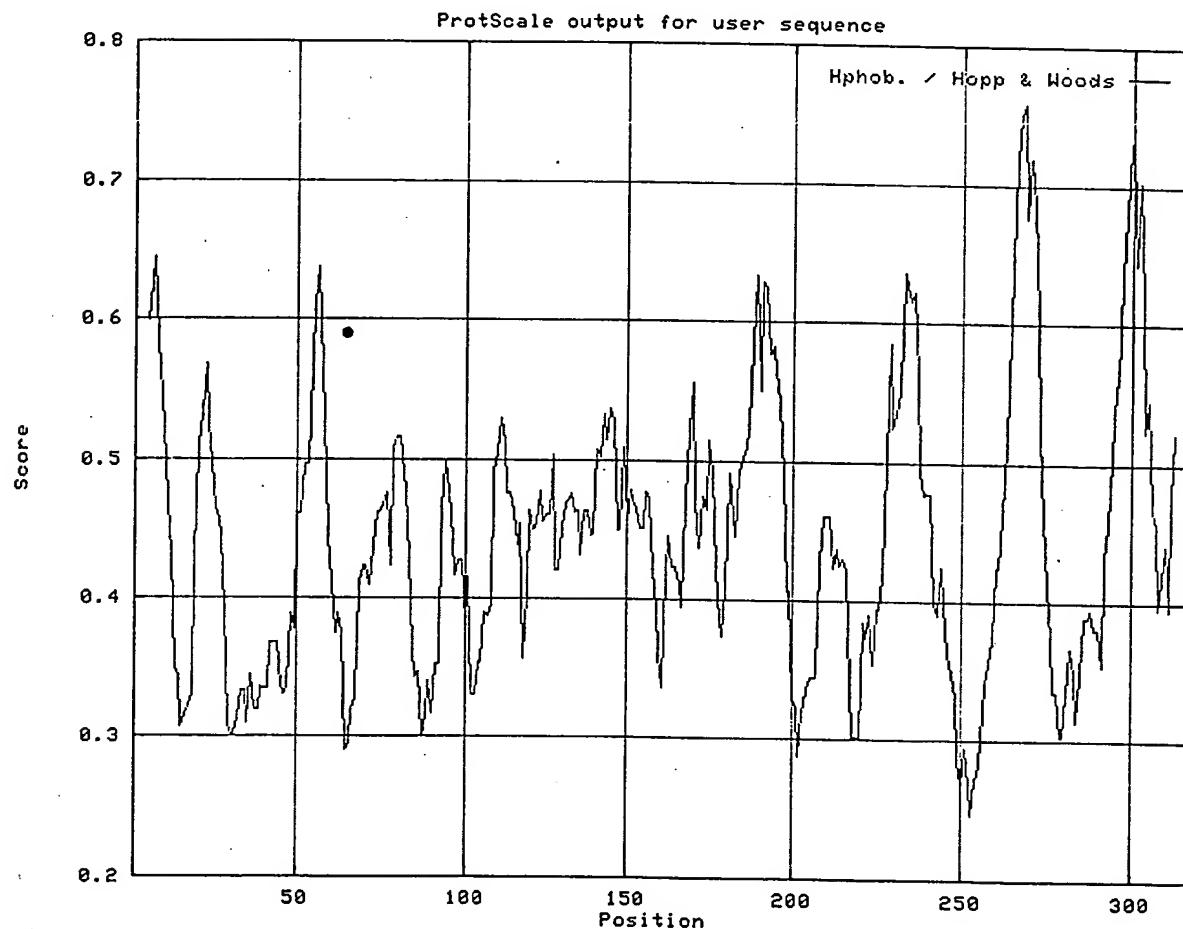


Figure 6:
101P3A11 Hydropathicity Profile
(Kyte J., Doolittle R.F., 1982. J. Mol. Biol. 157:105-132)

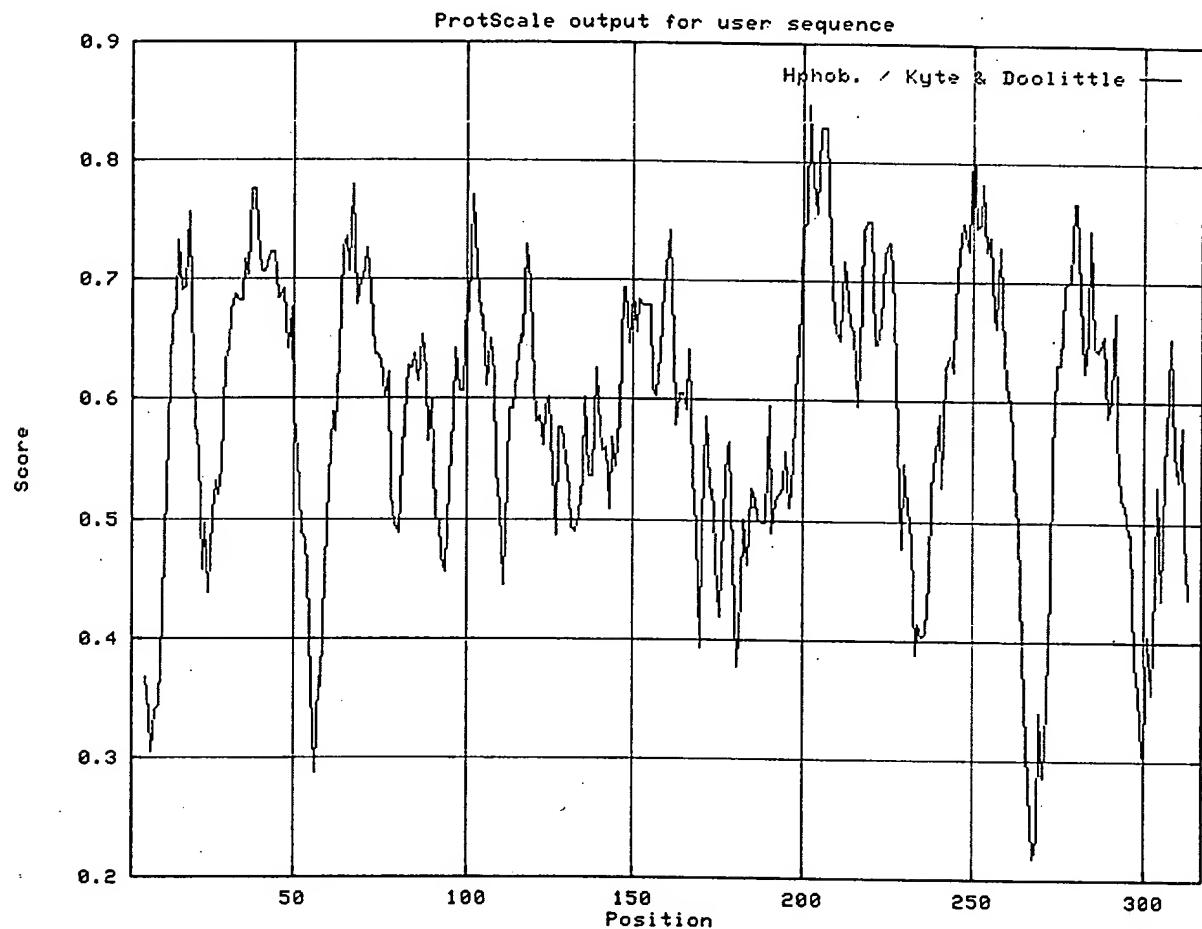


Figure 7:
101P3A11 % Accessible Residues Profile
(Janin J., 1979. Nature 277:491-492)

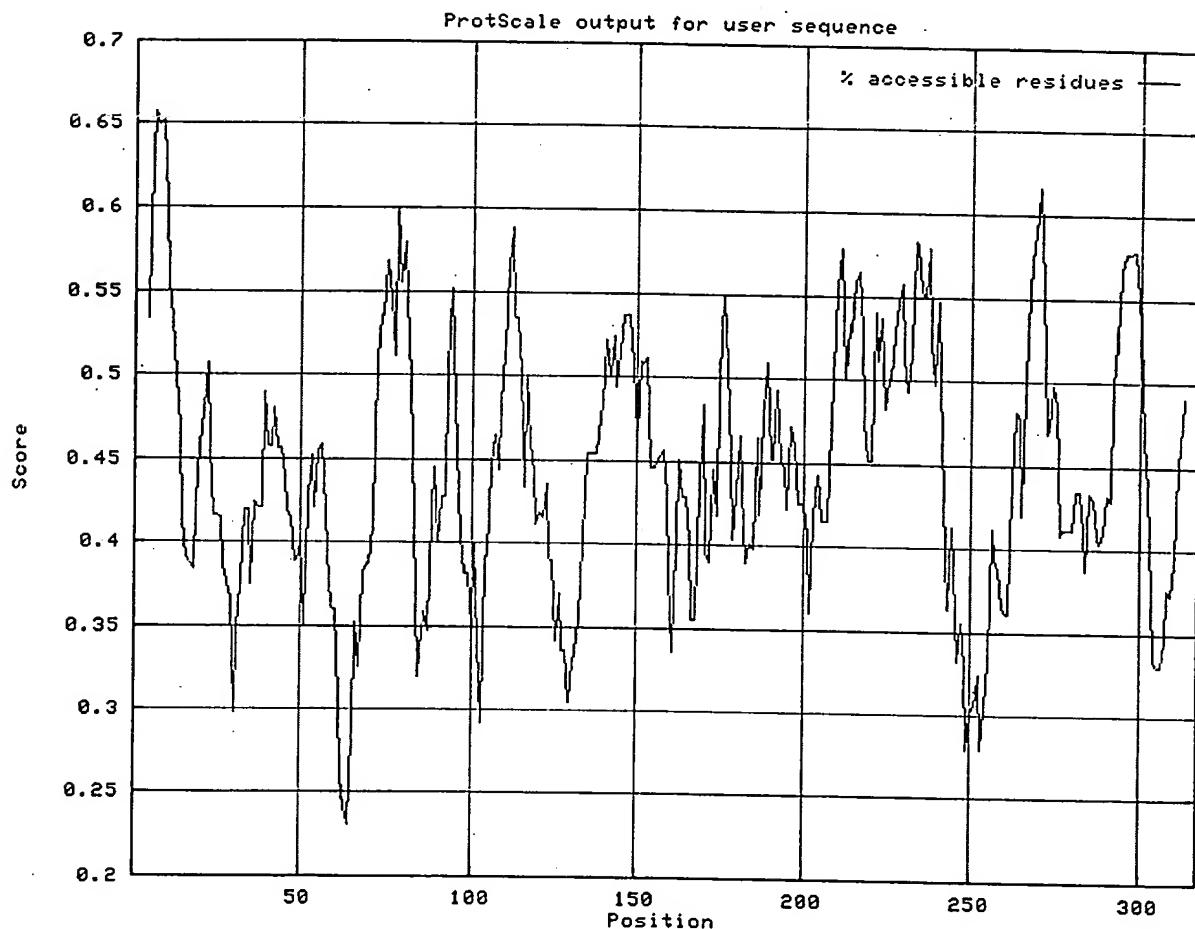


Figure 8:
101P3A11 Average Flexibility Profile
(Bhaskaran R., Ponnuswamy P.K., 1988.
Int. J. Pept. Protein Res. 32:242-255)

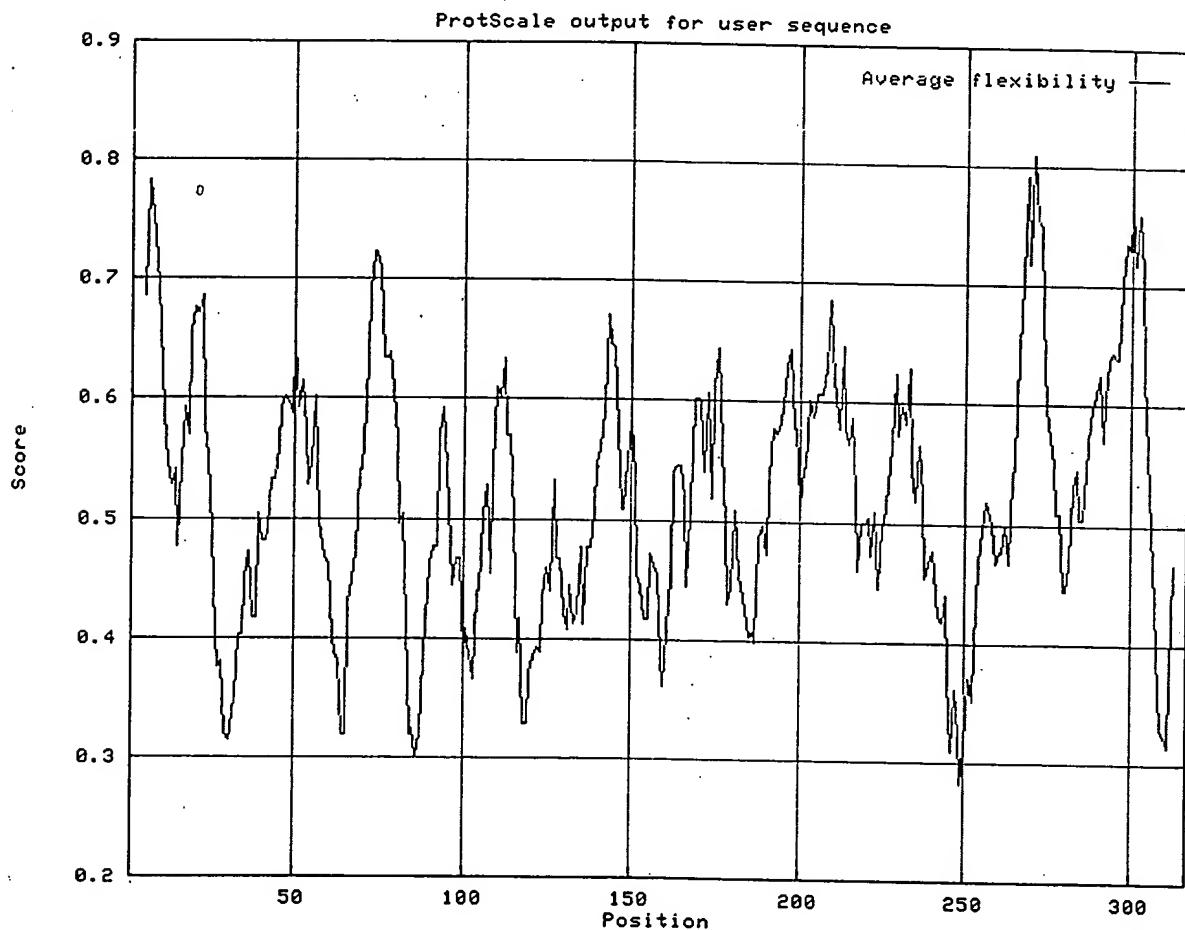


Figure 9:
101P3A11 Beta-turn Profile
(Deleage, G., Roux B. 1987. Protein Engineering 1:289-294)

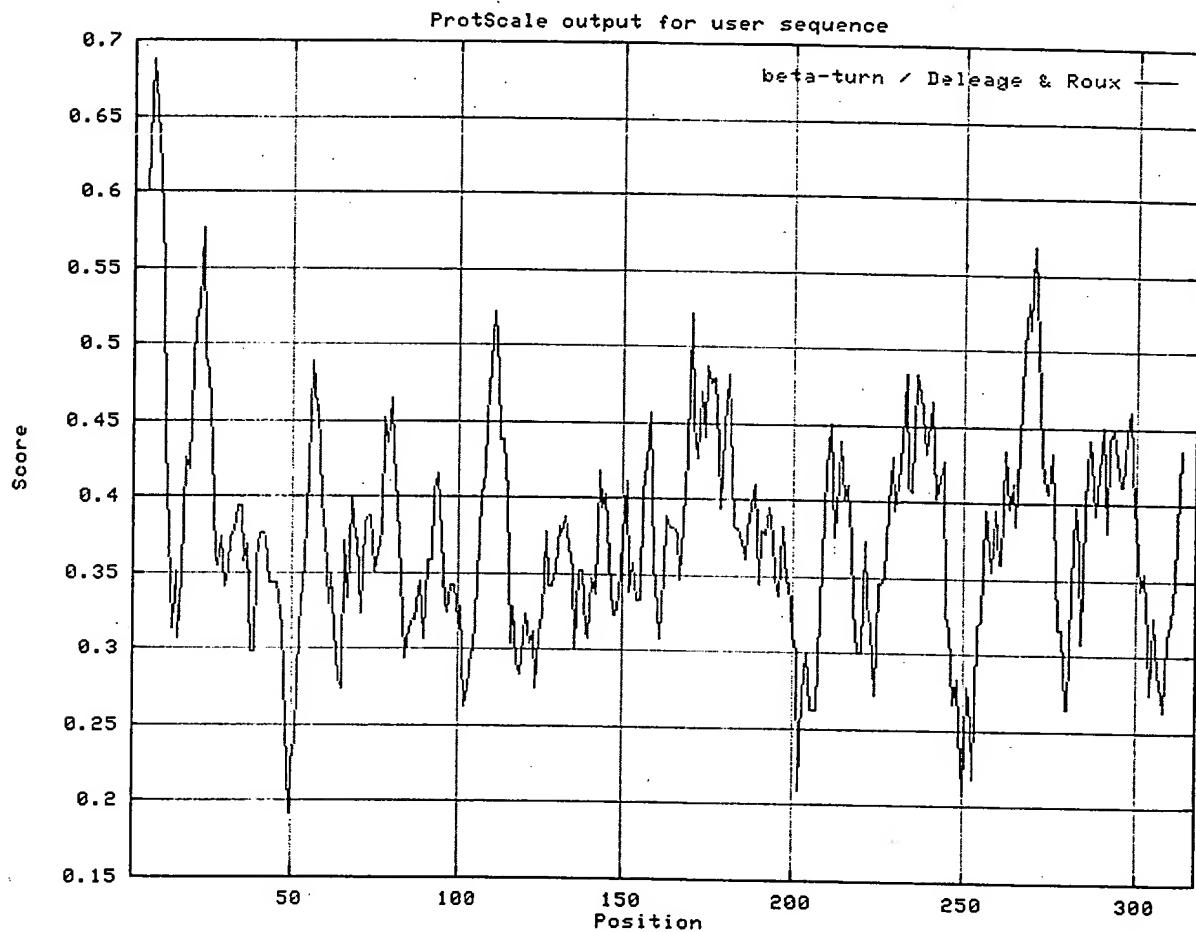
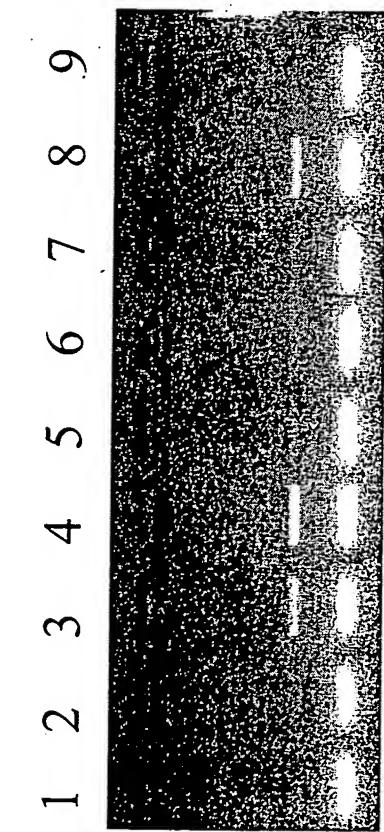




Figure 10A. Expression of 101P3A11 by RT-PCR



- VP1 (Kidney, Lung, Liver)
- VP2 (Pancreas, Colon, Stomach)
- Prostate xenograft Pool
- Prostate Cancer Pool
- Kidney Cancer Pool
- Colon Cancer Pool
- Breast Cancer Pool
- Metastasis Pool
- H₂O

Figure 10B

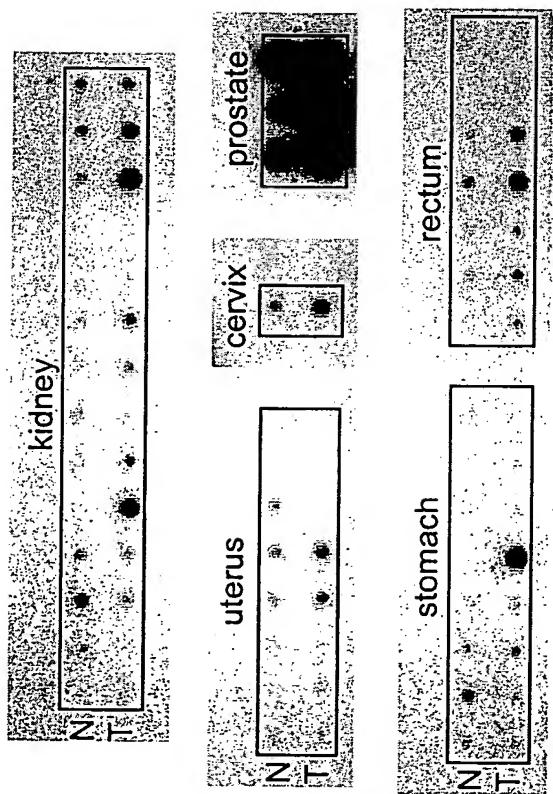




Figure 11. Expression of 101P3A11 in Human Patient Cancer Specimens

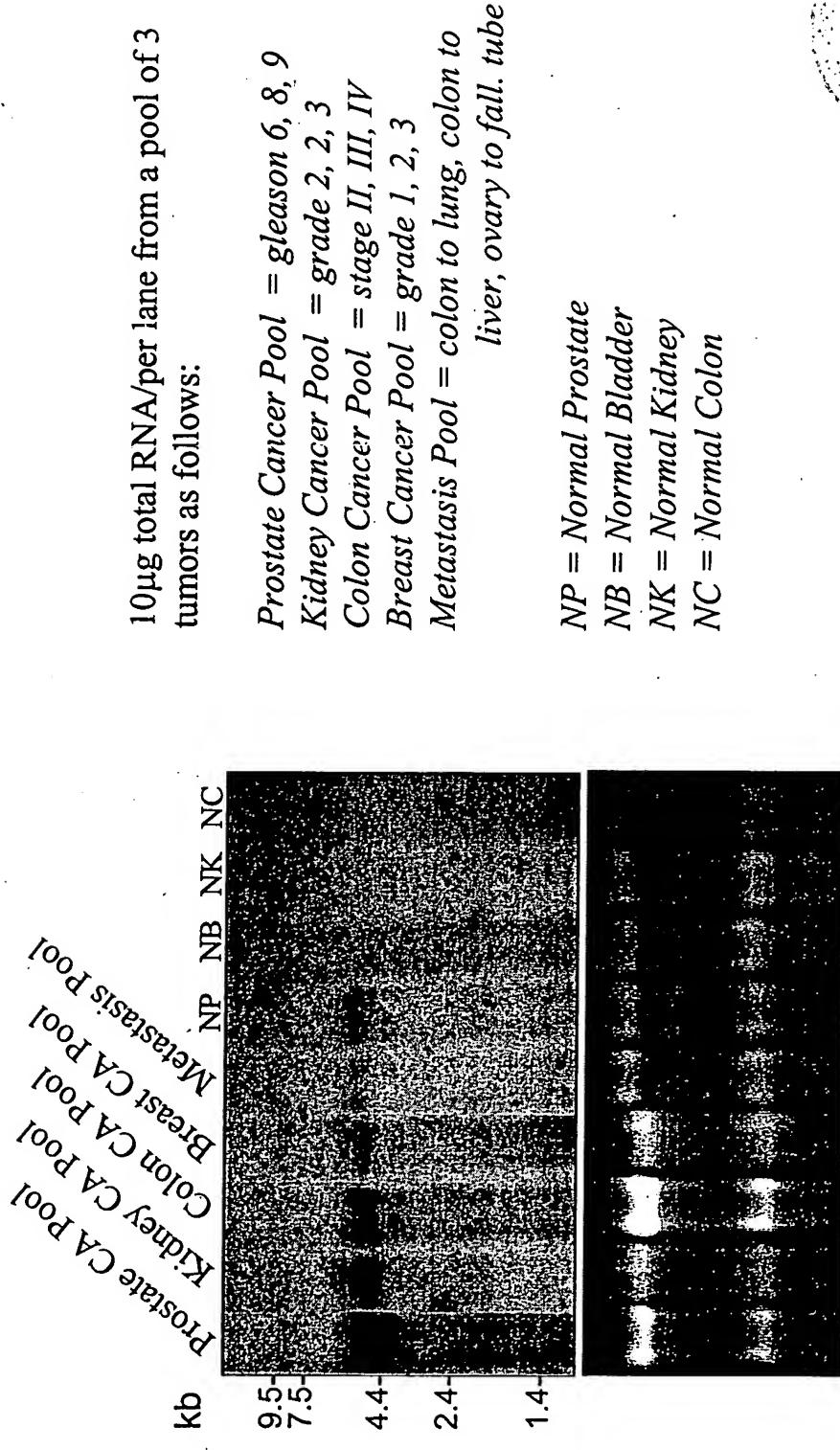


Figure 12A

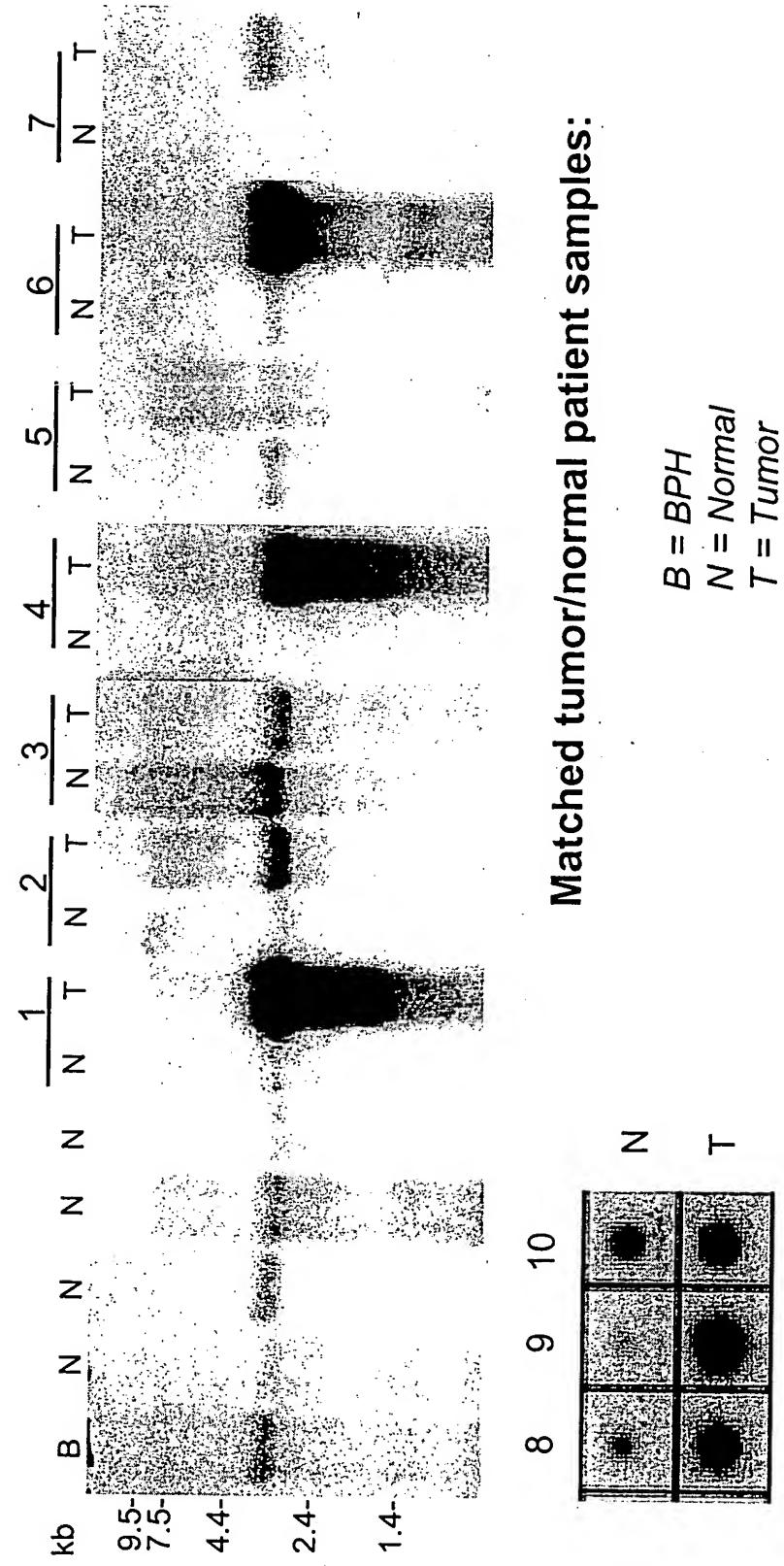


Figure 12B and 12C

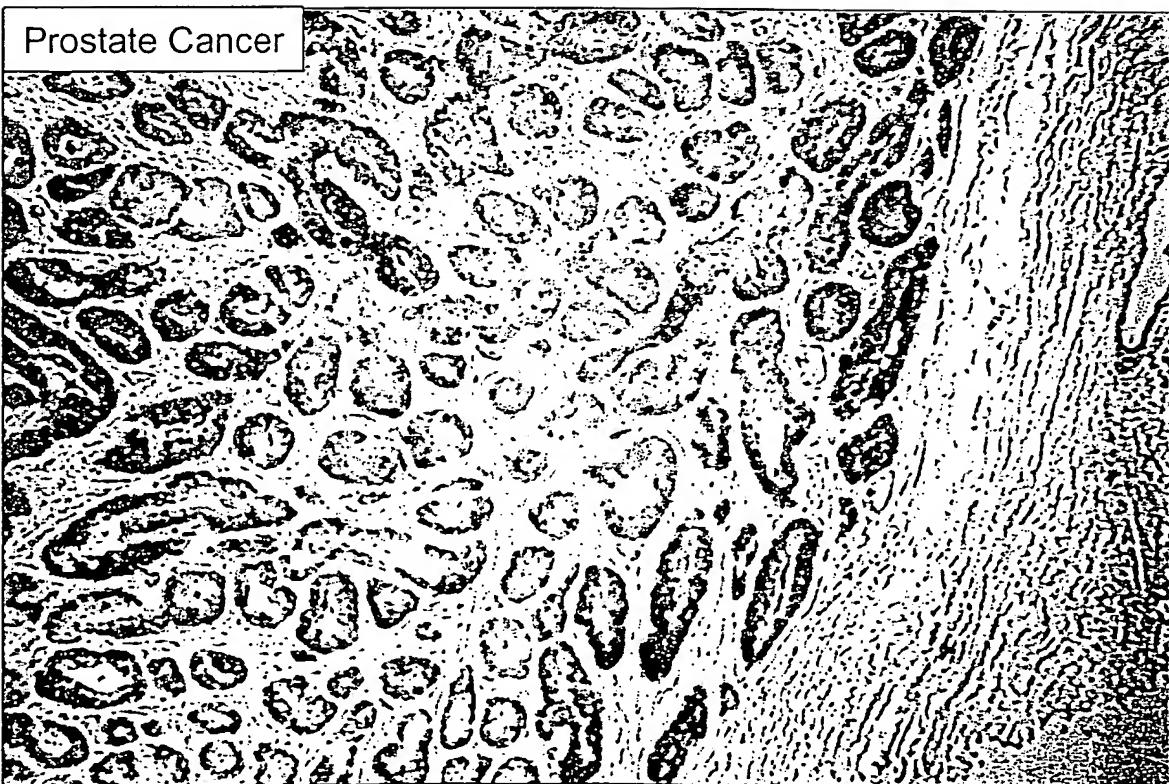
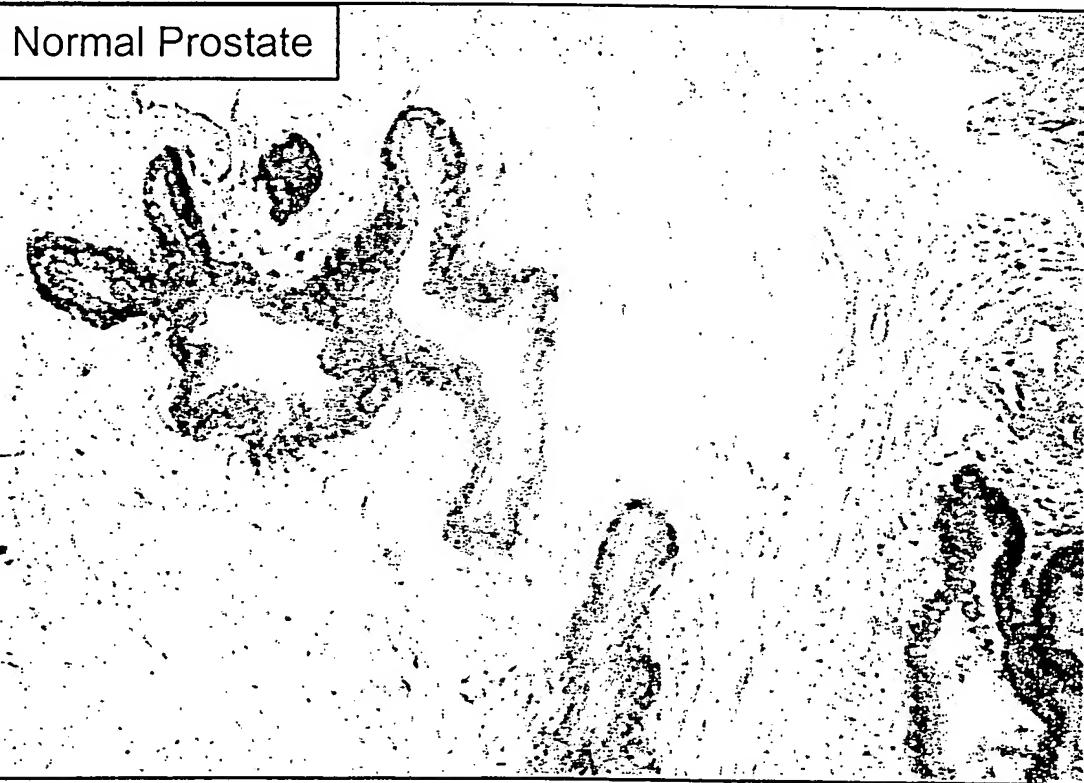
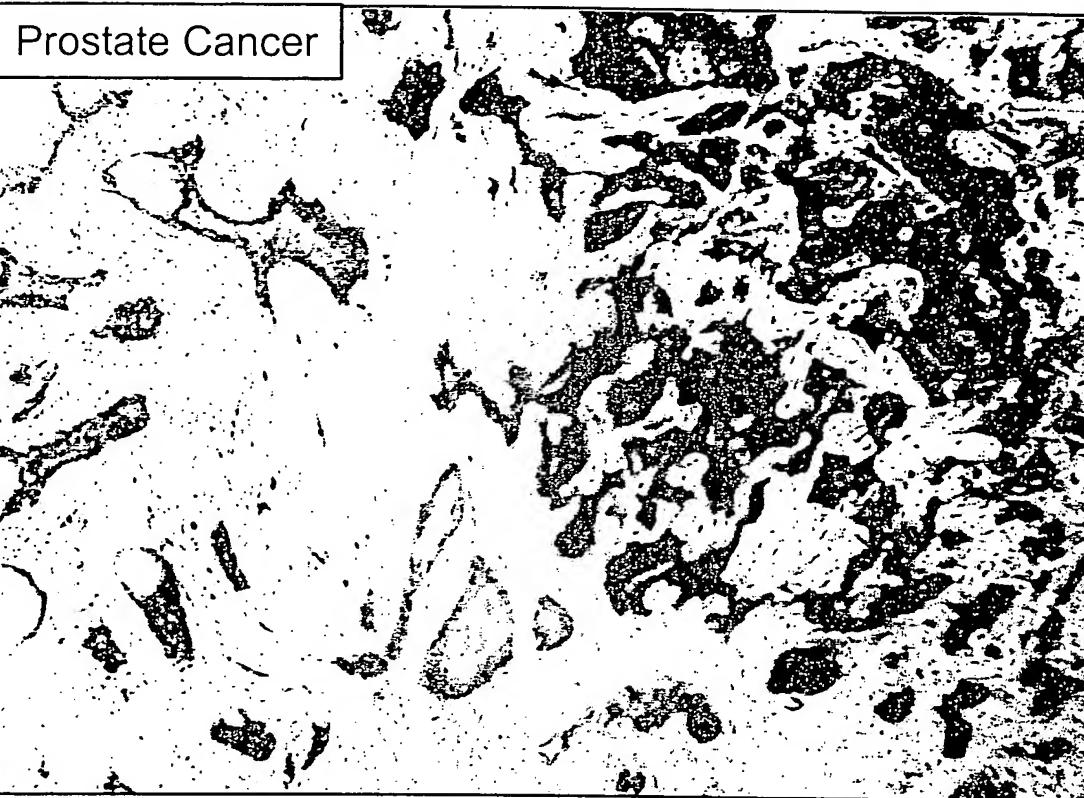


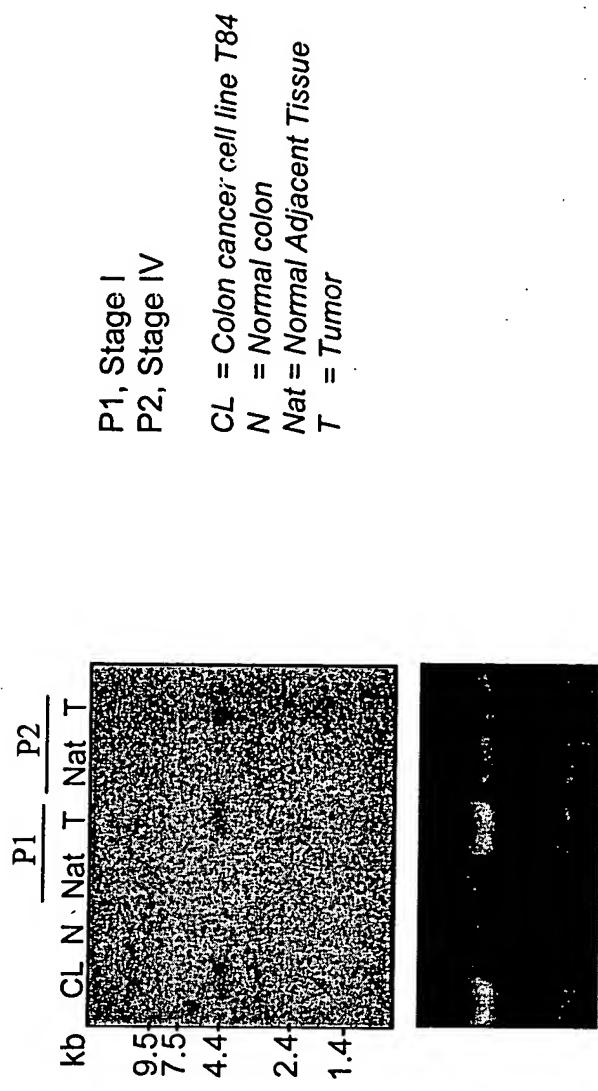
Figure 12D and 12E



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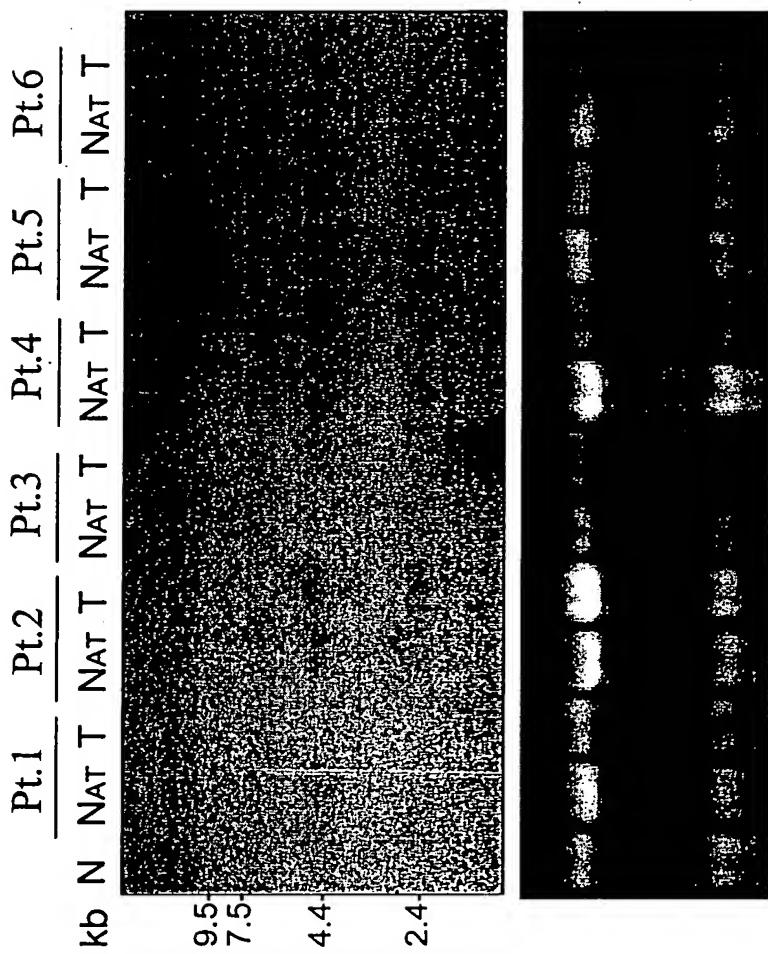


Figure 13. Expression of 101P3A11 in Colon Cancer Patient Specimens



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Figure 14. Expression of 101P3A11 in Kidney Cancer Patient Specimens



Pt.1, Papillary carcinoma, stage I
Pt.2, Invasive papillary carcinoma
Pt.3, Clear cell type grade 1/3, focally 2/3
Pt.4, Clear cell type, stage III
Pt.5, Clear cell type, stage III
Pt.6, Clear cell type, stage III

N = Normal kidney

NAT = Normal adjacent tumor

T = Tumor

Figure 15A-15C. Androgen Regulation of 101P3A11 in Tissue Culture Cells

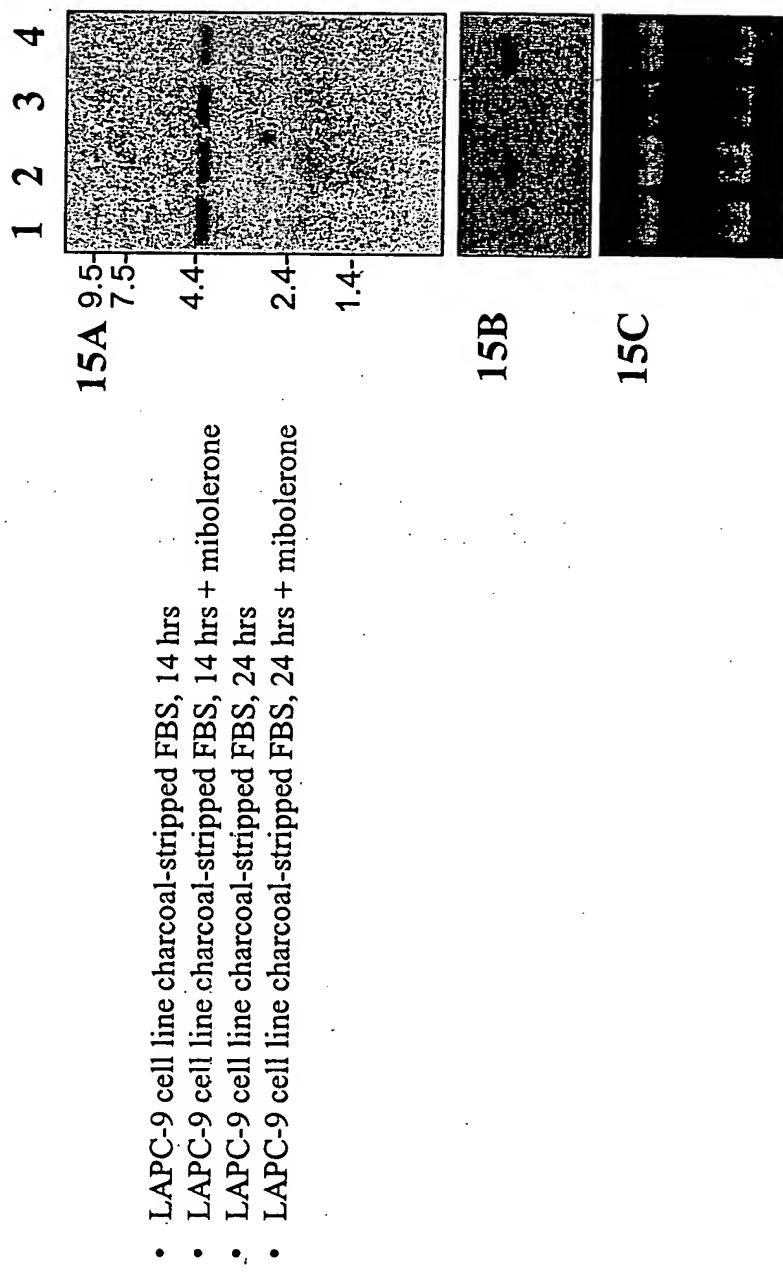
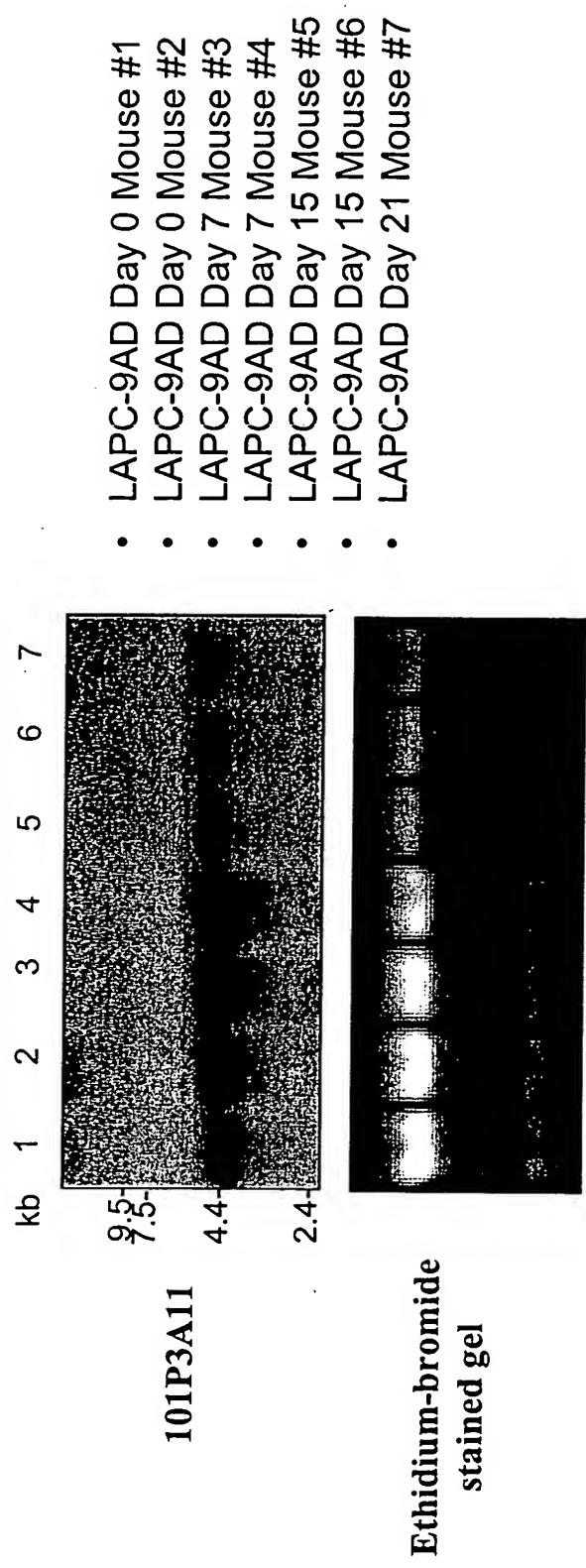
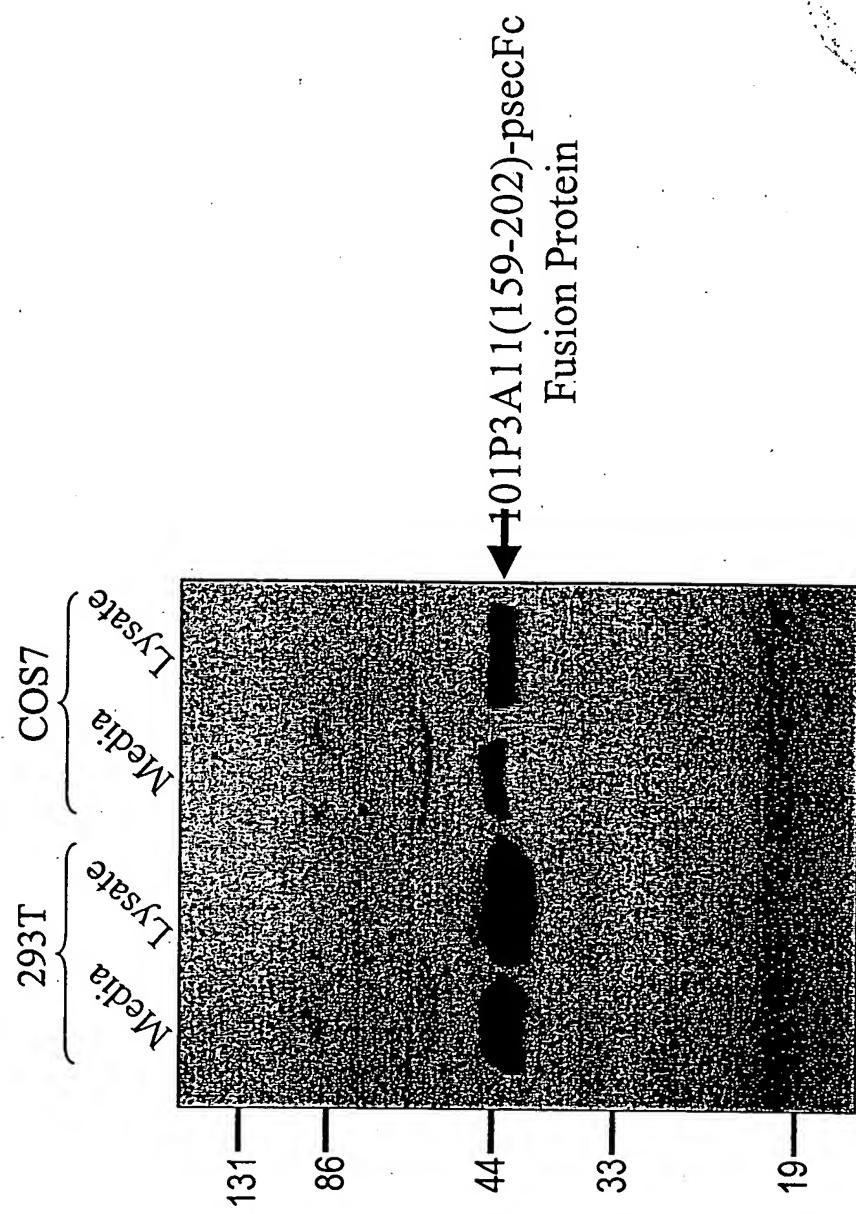


Figure 16. Androgen Regulation of 101P3A11 *In Vivo*



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Figure 17. Expression and Detection of 101P3A11(159-202)-psecFc Fusion Protein



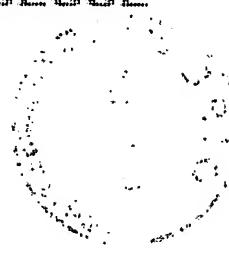
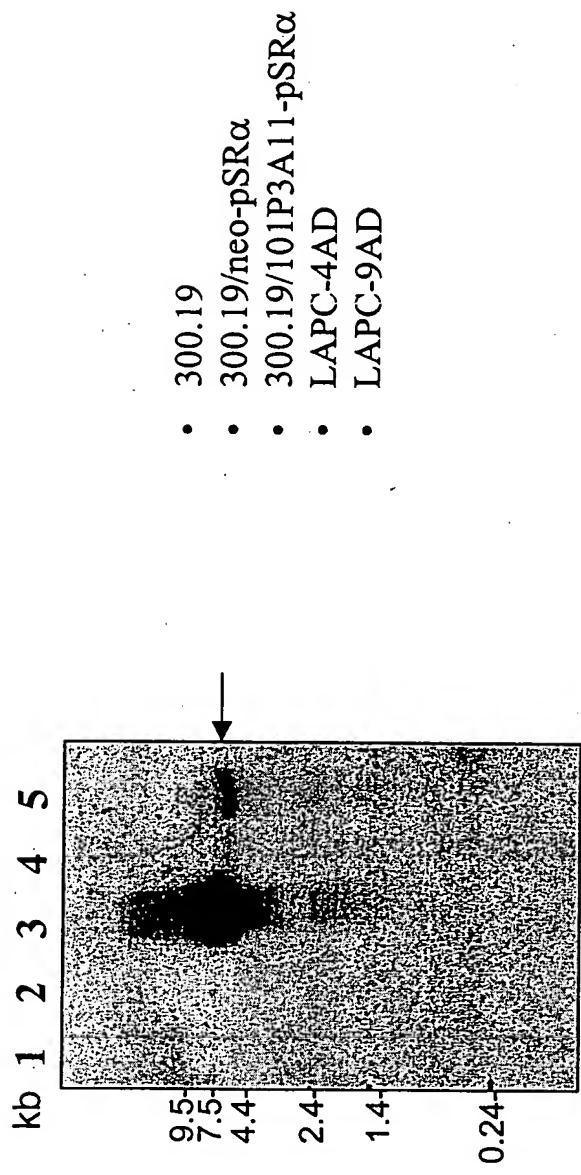


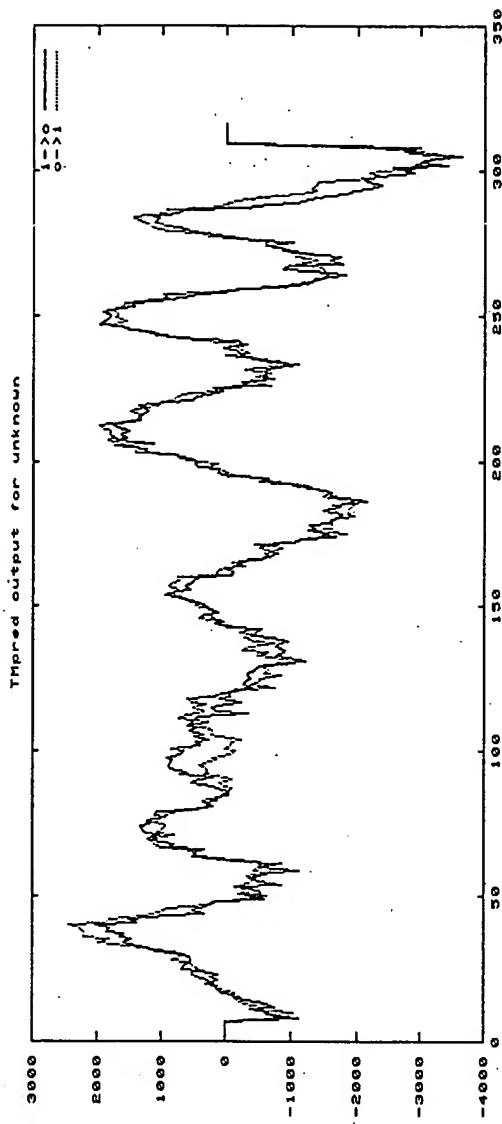
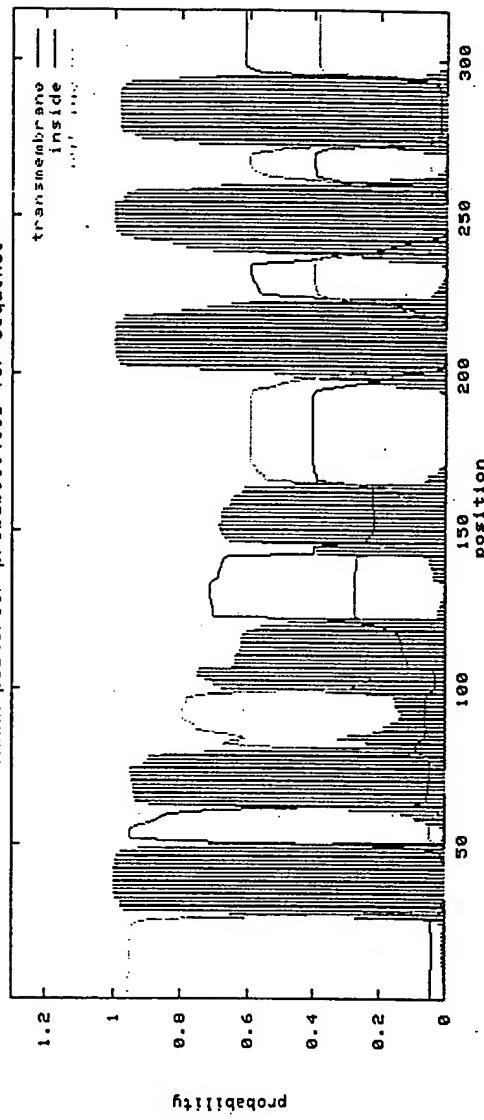
Figure 18. Expression of 101P3A11 in 300.19 Cells



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Figure 19A: Secondary structure prediction of 101P3A11

10	20	30	40	50	60	70
MVDPNGNESSATYFILIGPLGLEEAQFWLAFPLCSLYLIAVLGNLTIIYIVRTEHSLHEPMYIFLCMLSG cccccccccceeeeeeccccchhhhhhhhhcchhhhhhhccceeeeeeccccchhhhhhhhhcc IDLISTSSMPKMLAIFWFNSTTIIQFDACLLQIAHSLSGMESTVILLAMAFDRYVAICHPLRHATVTL ceeeeeccchhhheeeeeccccccehhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhhh PRVTKIGVAAVVRGAALMAPLPVFIKQLPFCRSNITLSHSYCLHQDVMKIACDDIRVNVVYGLIVIISAG cccheehhhhhhhhhccchhhheccccccchhhccchhhhhhhhhhhhhhhhhhhhh LDSILLISFSYLLILKTVLGLTREAQAKAFGTCVSHVCASFIFYVPFIGLSMVRFSKRARDSPILPVILANI hh YLVPPVLPNPIVYGVKTKEIRQRILRLFHVATHASEP hhhhccccccchheeechhhhhhhhhhhhhhhhhhhhhhh						

Figure 19B-19C. Transmembrane prediction of 101P3A11**19B****19C**

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Figure 20. Expression of 101P3A11 in NIH-3T3 Tumors

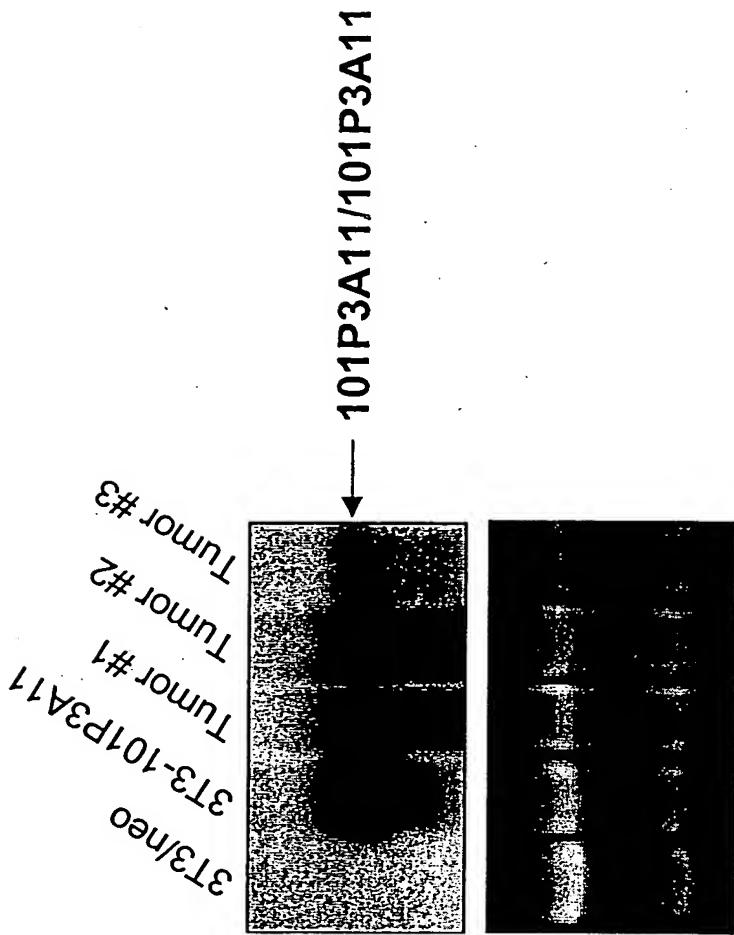
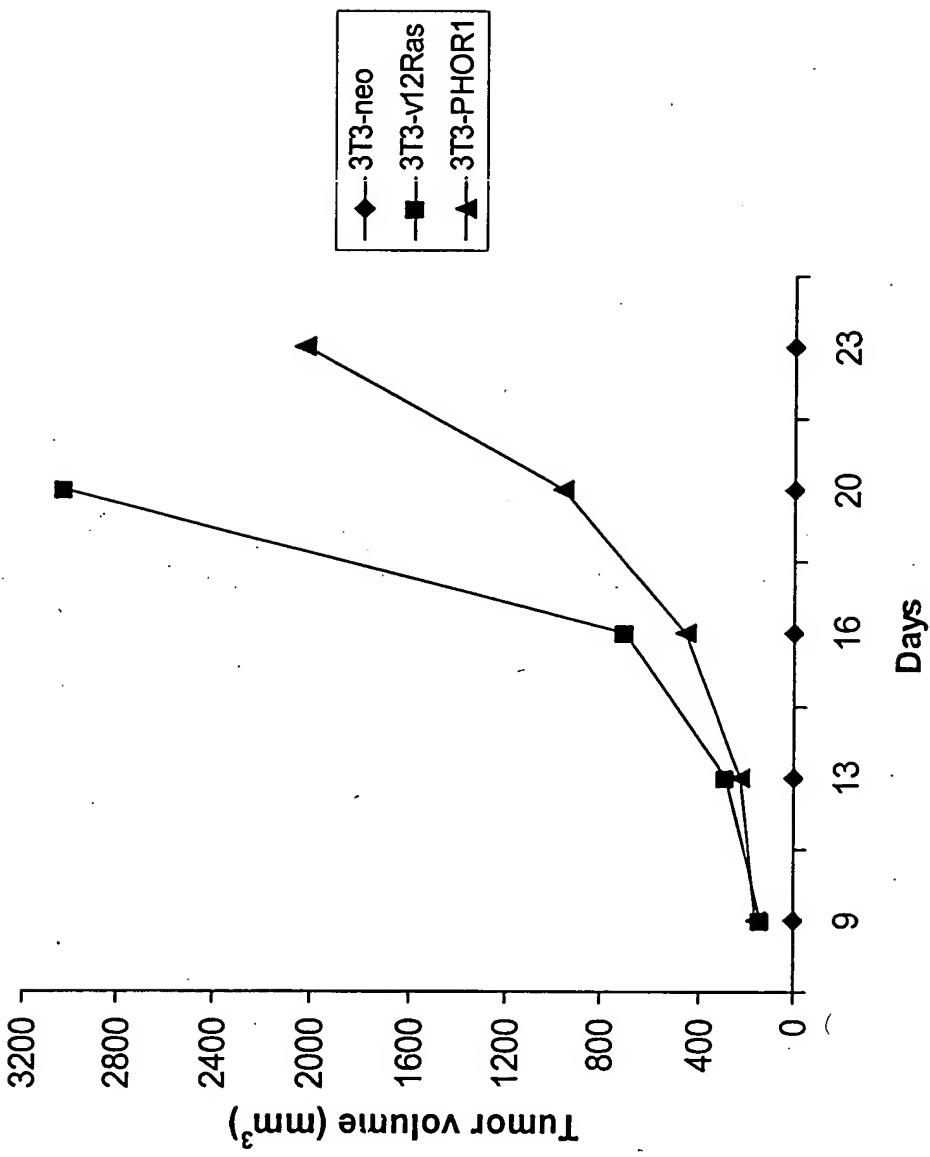
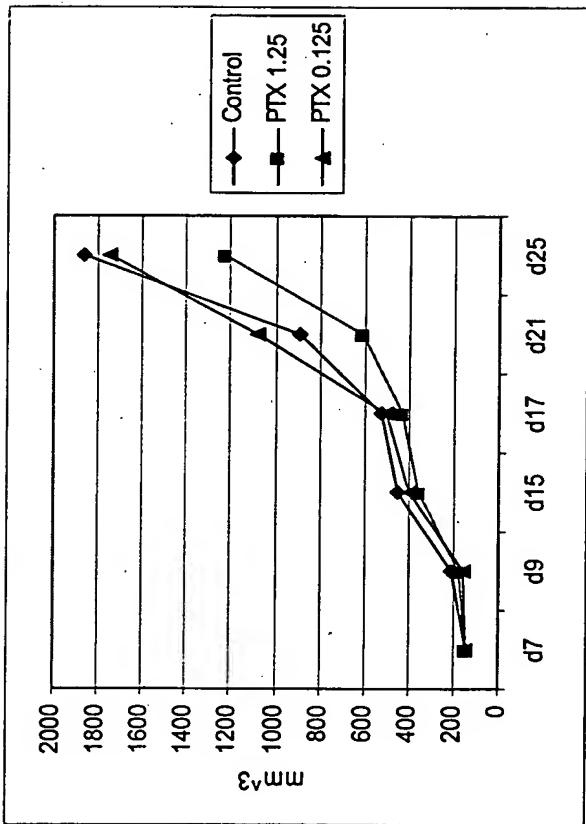


Figure 21: 101P3A11 Induces Tumor Formation of 3T3 Cells



• Injection of 10^6 3T3-neo, 3T3-Ras or 3T3-101P3A11 cells subcutaneously into SCID mice revealed that 6/6 3T3-Ras-injected mice formed tumors, 6/6 3T3-101P3A11-injected mice formed tumors, and 0/6 3T3-neo-injected mice formed tumors.

Figure 22: PTX Reduces the *in vivo* Growth of 3T3-101P3A11 Tumors



- Pertussis toxin inhibits the sub-cutaneous growth of 3T3-101P3A11 tumors in SCID mice.
- The inhibitory activity of pertussis toxin occurs in a dose dependent manner.

Figure 23: Alignment of 101P3A11-PHOR-1 with the rat GPCR RA1C (gi|3420759).

Identities = 179/299 (59%), Positives = 231/299 (76%), Gaps = 1/299 (0%)

PHOR: 14	FILIGLPGLEEAQFWLAFLPLCSLYLIAVLGNLTIIYIVRTEHSLHEPMYIFLCMLSGIDI	73
	F+LIG+PGLEEA FW FPL S+Y +A+ GN +++IVRTE SLH PMY+FLCML+ ID+	
RA1C: 11	FMLIGIPGLEEAHFWFGFPLLSMYAVALFGNCIVVIFVRTERSLHAPMYLFCLMLAAIDL	70
PHOR: 74	LISTSSMPKMLAIFWFNSTTIQFDACLLQIFAIHSLSGMESTVLLAMAFDRYVAICHPLR	133
	+STS+MPK+LA+FWF+S I FDACL Q+F IH+LS +EST+LLAMAFDRYVAICHPLR	
RA1C: 71	ALSTSTMPKILALFWFDSREITFDACLAQMFFIHALSAIESTILLAMAFDRYVAICHPLR	130
PHOR: 134	HATVLTLPRVTKIGVAAVVRAALMAPLPVFIKQLPFCRSNILSHSYCILHQDVMKLACDD	193
	HA VL +IG+ A+VRG+ PLP+ IK+L FC SN+LSHSYC+HQDVMKLA D	
RA1C: 131	HAAVLNNNTVTQIGMVALVRGSLFFFPLPLLIKRLAFCHSNVLSHSYCVHQDVMKLAYTD	190
PHOR: 194	IRVNVVYGLIVIIISAIGLDSLISFSYLLILKTVLGL-TREAQAKAFGTCVSHVCAVFIF	252
	NNVYGL I+ +G+D + IS SY LI++ VL L ++ +AKAFGTCVSH+ V F	
RA1C: 191	TLPNVVYGLTAILLVMGVDMFISLSYFLIRAVLQLPSKSERAKAFGTCVSHIGVVLA F	250
PHOR: 253	YVPFIGLSMVHRFSKRRDSPLPVILANIYLLVPPVLPPIVYGVKTKEIRQRILRLFHVA	311
	YVP IGLS+VHRF D + V++ ++YLL+PPV+NPI+YG KTK+IR R+L +F ++	
RA1C: 251	YVPLIGLSVVHRFGNSLDPIVHVLMGDVYLLLPPVINPIIYGAKTKQIRTRVLAMFKIS	309

Figure 24: Alignment of 101P3A11-PHOR-1 with the human prostate specific GPCR. (gi|13540539)

Identities = 179/299 (59%), Positives = 233/299 (77%), Gaps = 1/299 (0%)

PHOR: 14	FILIGLPGLEEAQFWLAFPLCSLYLIAVLGNLTIIYIVRTEHSLHEPMYIFLCMLSGIDI	73
	F+LIG+PGLE+A FW+ FPL S+Y++A+ GN +++IVRTE SLH PMY+FLCML+ ID+	
GPCR: 11	FVLIGIPGLEKAHFVWGFPLLSMYVVAMFGNCIVVFIVTERSLHAPMYLFLCMLAAIDL	70
PHOR: 74	LISISSLMPKMLAIFWFNSTTIQFDACLLQIFAIHSLGMESTVLLAMAFDRYVAICHPLR	133
	+STS+MPK+LA+FWF-S I F+ACL Q+F IH+LS +EST+LLAMAFDRYVAICHPLR	
GPCR: 71	ALSTSTMPKILALFWFDSREISFEACLTQMFFIHALSAIESTILLAMAFDRYVAICHPLR	130
PHOR: 134	HATVLTLPRVTKIGVA\VVVRGAALMAPLPVFIKQLPFCRSNILSHSYCLHQDVMKLACDD	193
	HA VL +IG+ AVVRG+ PLP+ IK+L FC SN+LSHSYC+HQDVMKLA D	
GPCR: 131	HAAVILNNNTVTAQIGIVAVVRGSLFFFPLPLLKRLAFCHSNVLSHSYCVHQDVMKLAYAD	190
PHOR: 194	IRVNVVYGLIVIISAIGLDSLISFSYLLILKTVLGL-TREAQAKAFGTCVSHCAVFIF	252
	NVYGL I+ +G+D + IS SY LI++TVL L ++ +AKAFGTCVSH+ V F	
GPCR: 191	TLPNVVYGLTAILLVMGVDMFISLSYFLIIRTVLQLPSKSERAKAFGTCVSHIGVVLAF	250
PHOR: 253	YVPFIGLSMVHRFSKRRDSPLPVILANIYLLVPPVLPNPIVYGVKTKEIRQRILRLFVA	311
	YVP IGLS+VHRF + V++ +IYLL+PPV+NPI+YG KTK+IR R+L +F ++	
GPCR: 251	YVPLIGLSVVHRFGNSLHPIVRVVMGDIYLLLPPVINPIIYGAKTKQIRTRVLAMFKIS	309

Figure 25: Alignment with human olfactory receptor 5I12 (gi|14423836)

Identities = 163/304 (53%), Positives = 214/304 (69%), Gaps = 1/304 (0%)

PHOR: 7 NESSATYFILIGLPGLEEAQFWLAFLPLCSLYLIAVLGNLTIIYIVRTEHSLHEPMYIFLC 66
N + +F+L G+PGL + WL+ PLC +Y +A+ GN I+ VR E SLHEPMY FL

HOR5: 5 NVTHPAFFLLTGIPGLESSHWSLGPLCVMYAVALGGNTVILQAVRVEPSLHEPMYYFLS 64

PHOR: 67 MLSGIDILISTSSMPKMLAIFWFNSTTIQFDACLLQIFAIHSLGMESTVLLAMAFDRYV 126
MLS D+ IS +++P +L F N+ I FDACL+Q+F IH S MES +LLAM+FDRYV

HOR5: 65 MLSFSDVAISMATLPTVLRTFCLNARNITFDACLIQMFLIHFFSMESGILLAMSFDRYV 124

PHOR: 127 AICHPLRHATVLTLPRTVKIGVAAVVRGAALMAPLPFIKQLPFCRSNI LSHSYCLHQDV 186
AIC PLR+ATVLT + +G+ A R + PLP IK+LP CRSN+LSHSYCLH D+

HOR5: 125 AICDPLRYATVLTTEVIAAMGLGAAARSFITLFPLPFLIKRLPICRSNVLSHSYCLHPDM 184

PHOR: 187 MKLACDDIRVNVVYGLIVIIISAIGLDSLLISFSYLLILKTVLGL-TREAQAKAFGTCVSH 245
M+LAC DI +N +YGL V++S G+D I SY+LIL++V+ +RE + KA TCVSH

HOR5: 185 MRLACADISINSIYGLFVLVSTFGMDLFFIFLSYVLIILRSVMATASREERLKALNTCVSH 244

PHOR: 246 VCAVFIFYVPFIGLSMVHRFSKRRDPLPVILANIYLLVPPVLNPIVYGVKTKEIRQRIL 305
+ AV FYVP IG+S VHRF K + V+++N+YL VPPVLPN++Y KTKEIR+ I

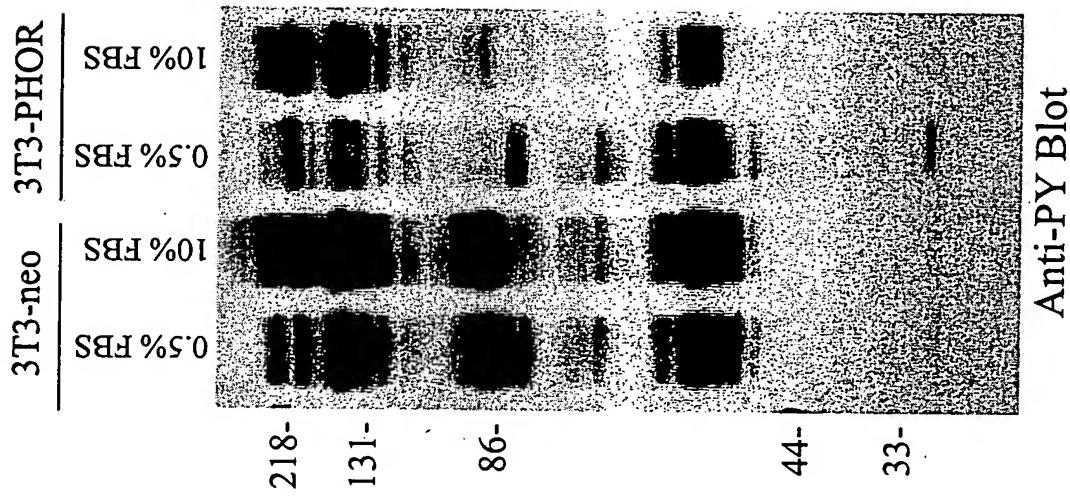
HOR5: 245 ILAVLAFYVPMIGVSTVHRFGKHVPCYIHVILMSNVYLFVPPVLNPLIYSAKTKEIRRAIF 304

PHOR: 306 RLFH 309

R+FH

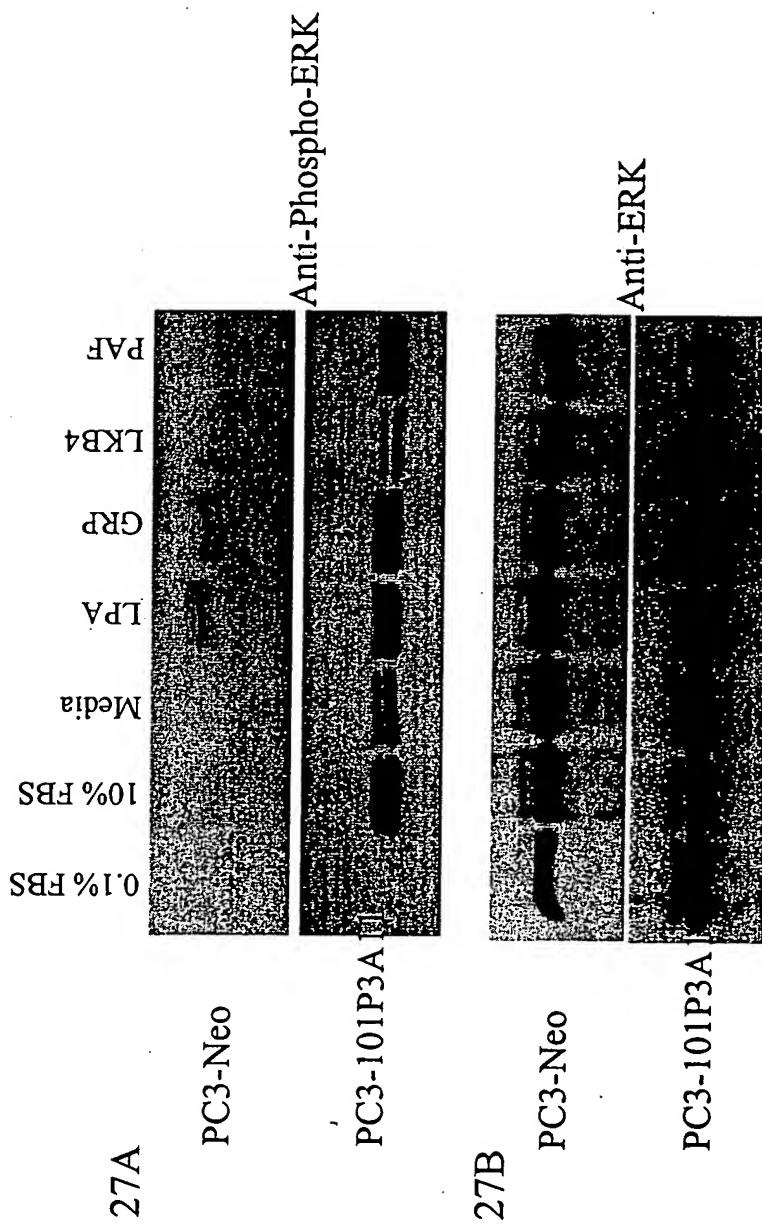
HOR5: 305 RMFH 308

Figure 26: 101P3A11 Modulated Tyrosine Phosphorylation in NIH-3T3 Cells



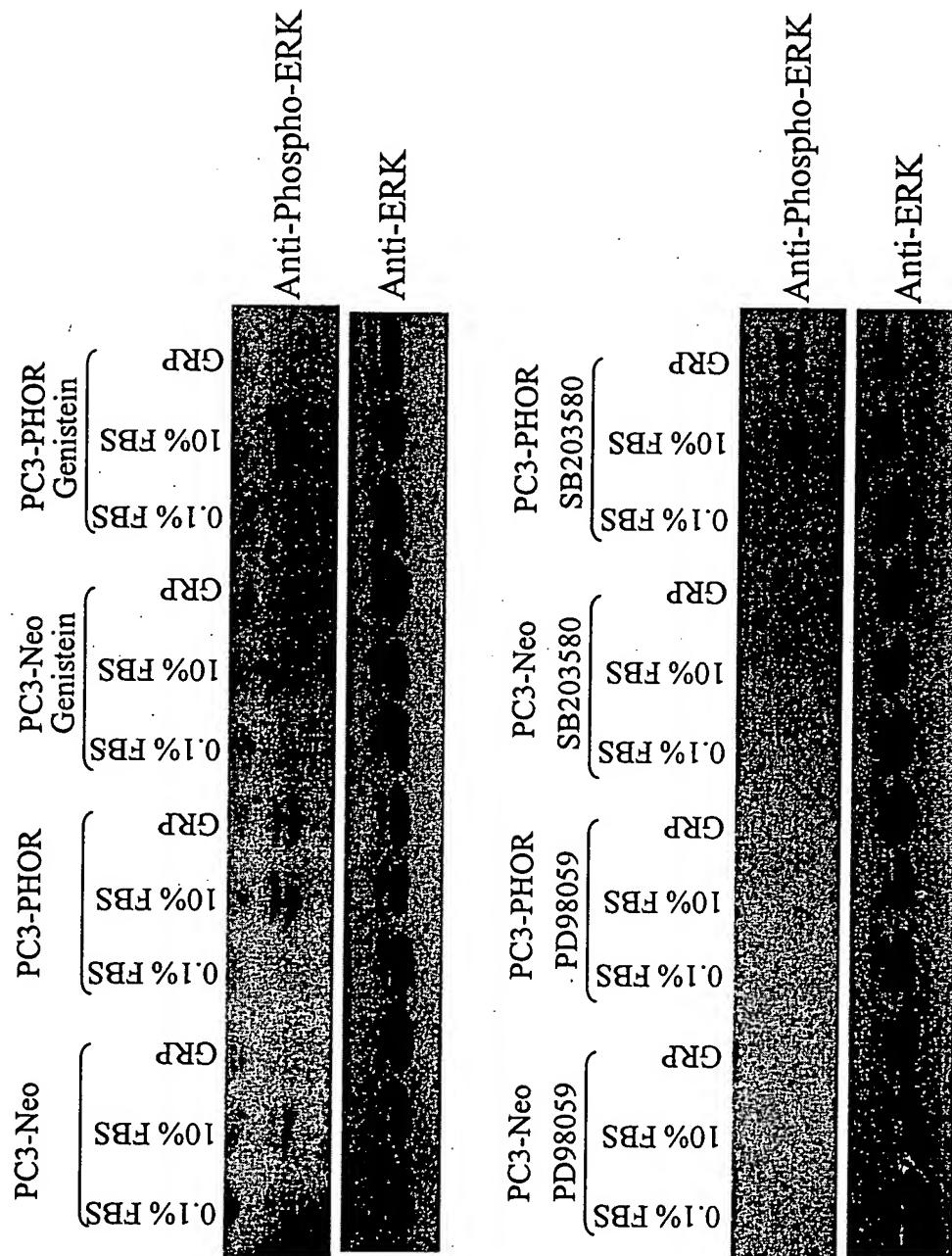
- 101P3A11 mediated the de-phosphorylation of proteins at 200, 120-140, 85-90 and 55 kDa.
- 101P3A11 induced the phosphorylation of proteins at 80 and 29 kDa in NIH-3T3 cells.

Figures 27A-27B: ERK Phosphorylation by PCR Ligands in 101P3A11 Expressing Cells



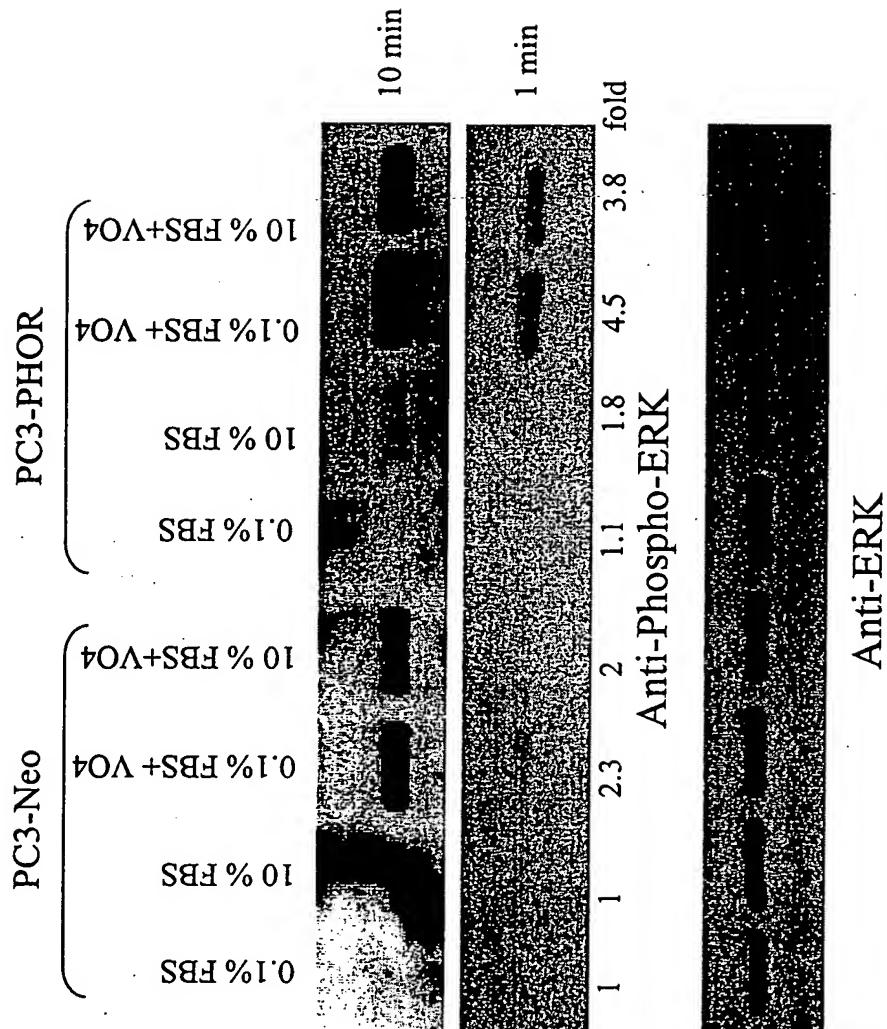
- FBS, lipophosphatidic acid, gastrin releasing peptide, leukotriene and platelet activating factor induced the phosphorylation of ERK in 101P3A11 expressing cells.

Figure 28: Inhibition of 101P3A11-Mediated ERK Activation by PD98059



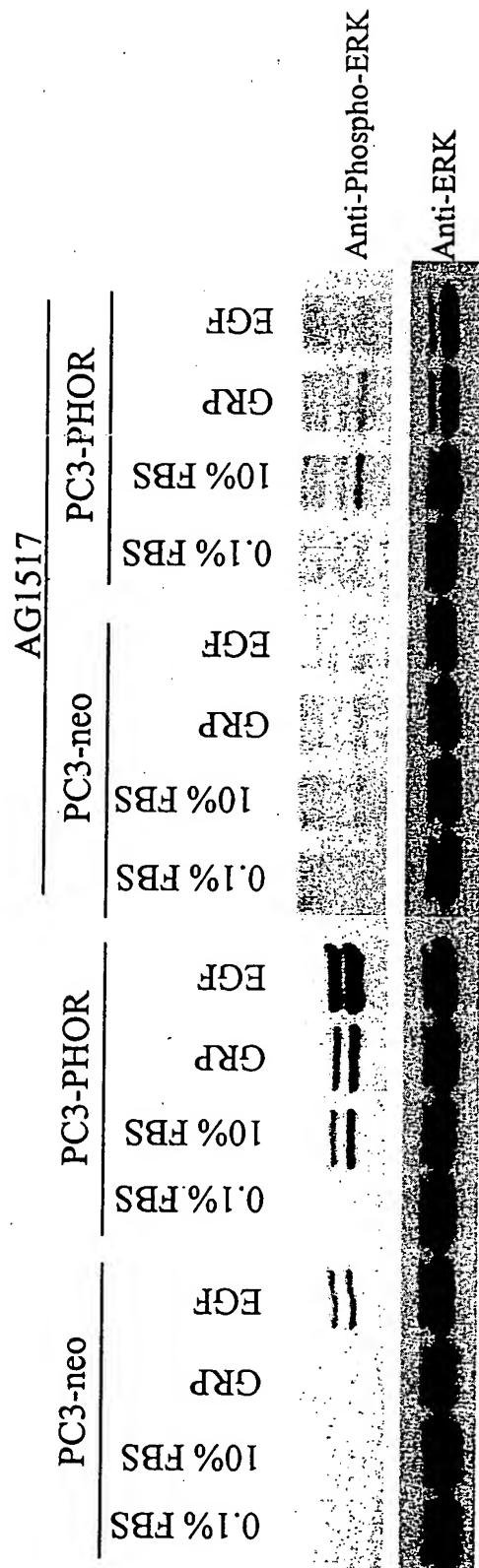
•ERK phosphorylation was inhibited by a MEK specific(PD98059) but not a p38 specific (SB203580) inhibitor in PC3-101P3A11 cells.

**Figure 29: Enhanced ERK Phosphorylation in Sodium Orthovanadate Treated
PC3-101P3A11 Cells**



•Sodium orthovanadate induced increased ERK phosphorylation in PC3-101P3A11 cells relative to PC3-neo cells.

Figure 30: Inhibition of 101P3A11-Mediated ERK Phosphorylation by AG1517



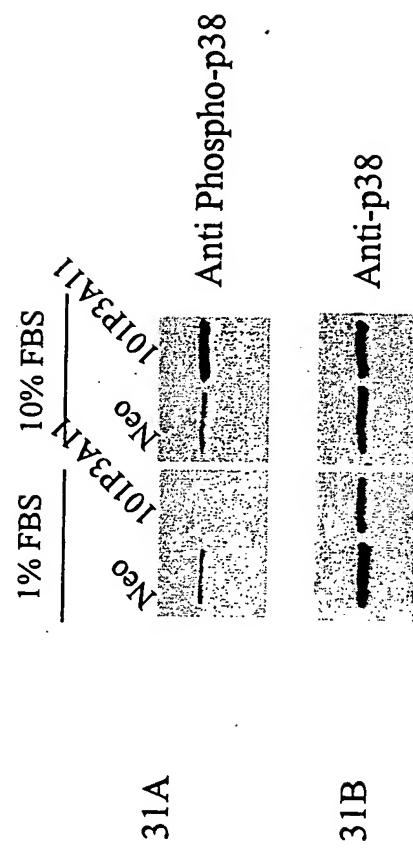
- The EGFR inhibitor, AG1517, inhibits EGF-mediated ERK phosphorylation in control and 101P3A11 expressing PC3 cells.
- AG1517 partially inhibits 101P3A11 mediated ERK phosphorylation in PC3 cells.

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Figure 31A-31B: Activation of p38 in PC3-101P3A11 Cells



- Expression of 101P3A11 mediates p38 phosphorylation in cells treated with 10% FBS.

Figure 32: 101P3A11 Induced Accumulation of cAMP in PC3 Cells

		Fold change in [cAMP]	
		PC3-Neo	PC3-PHOR
0.1%FBS	-PTX	1	4.302
	+PTX	1.403	2.577
10%FBS	-PTX	2.738	6.978
	+PTX	2.163	2.752

Fold Change in cAMP accumulation was calculated relative to PC3-neo cells grown in 0.1%FBS

- Expression of 101P3A11 increased the accumulation of cAMP in cells treated with 0.1% and 10% FBS.

- FBS-induced cAMP accumulation in 101P3A11 cells was inhibited by pertussis toxin.

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• Pertussis toxin inhibited FBS- mediated ERK phosphorylation in 101P3A11 expressing cells.

Figure 33: Pertussis Toxin Inhibits 101P3A11 Mediated ERK Phosphorylation

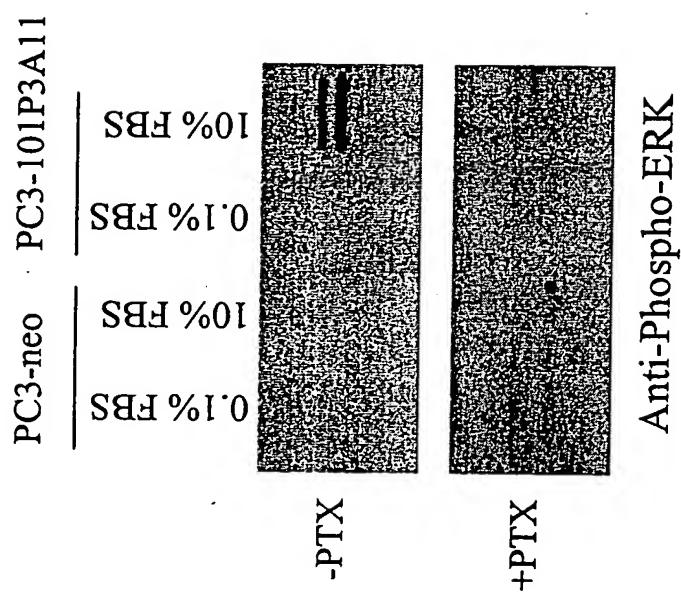
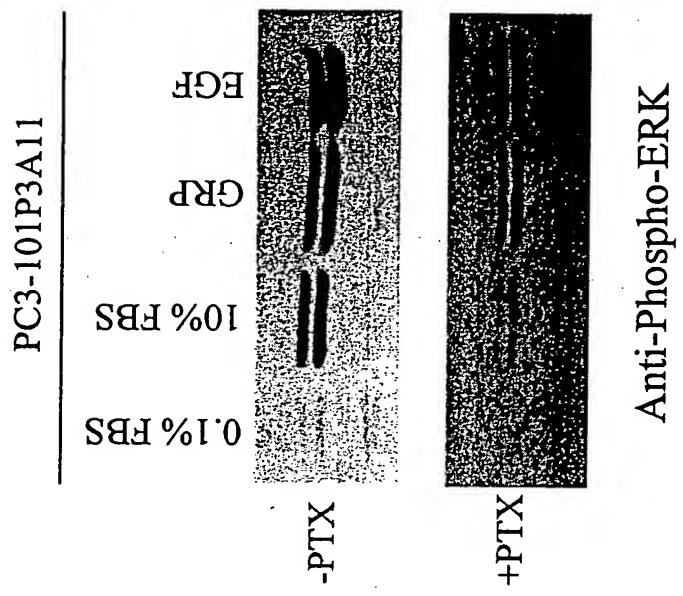


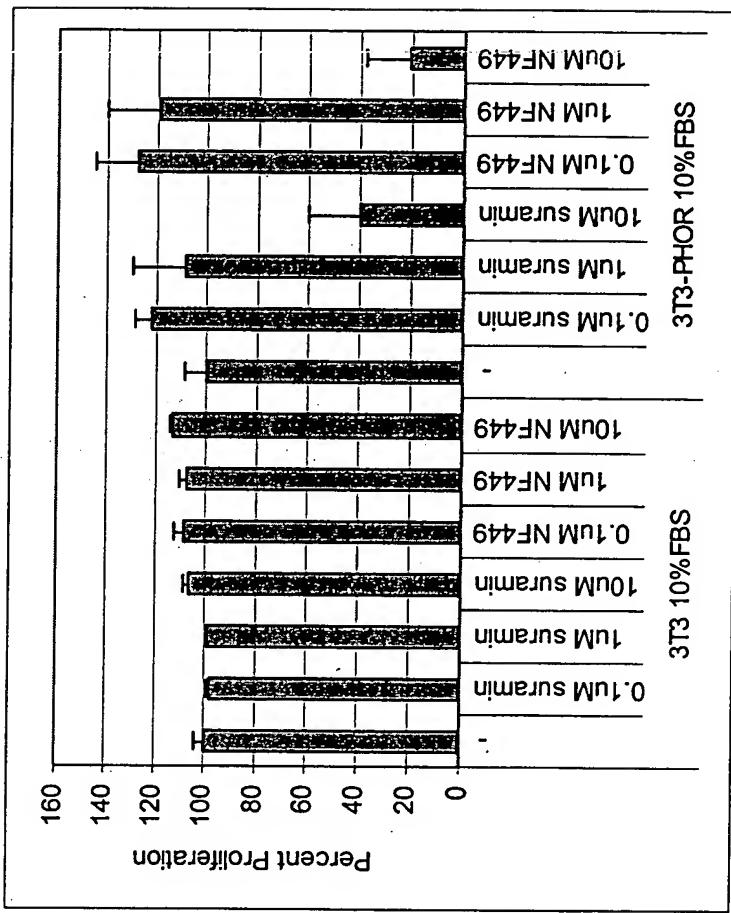
Figure 34: Pertussis Toxin Inhibited ERK Phosphorylation in PC3-101P3A11 Cells



• Pertussis toxin inhibited FBS- mediated ERK phosphorylation in 101P3A11 expressing cells.

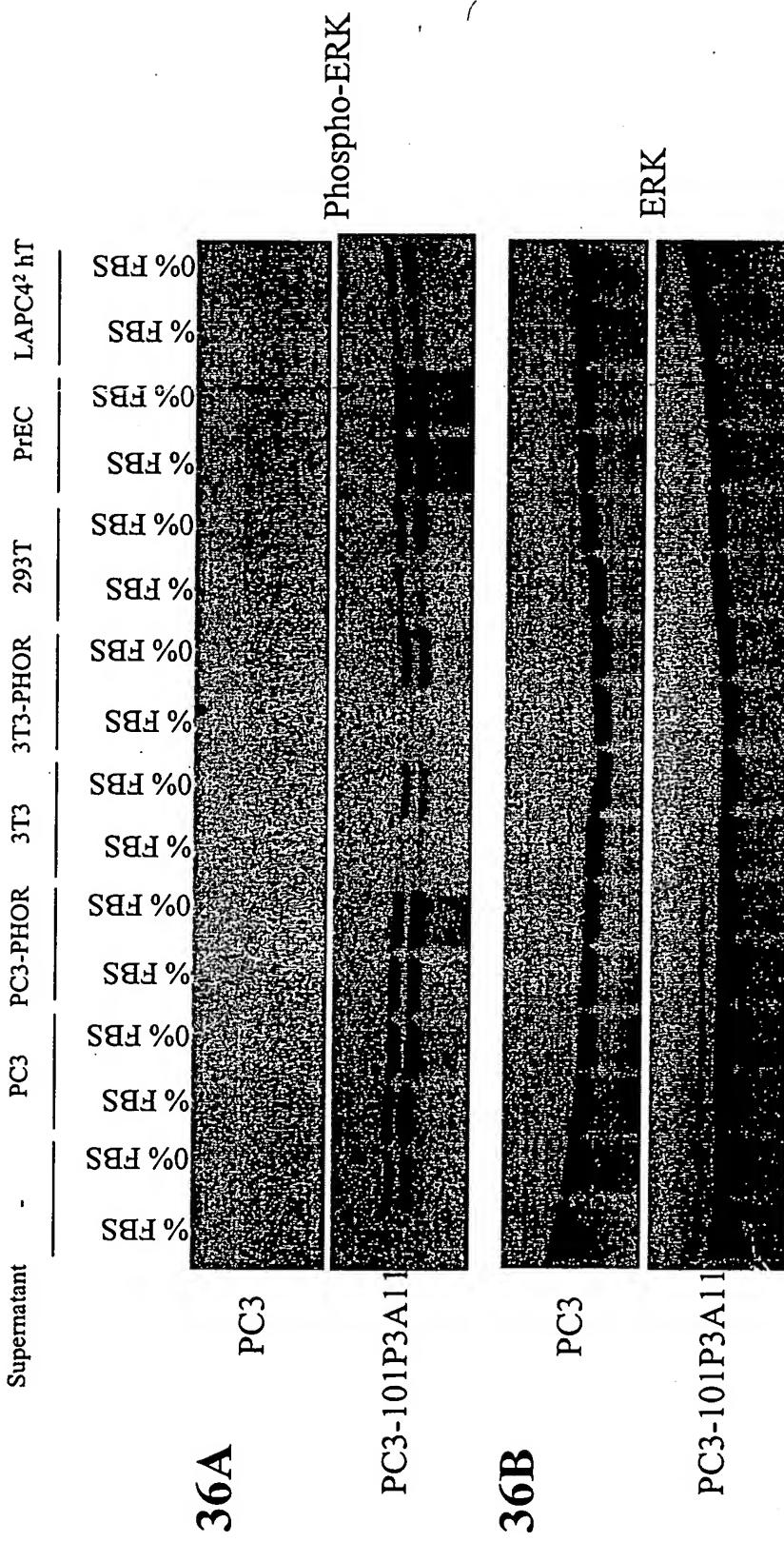
• The inhibitory activity of pertussis toxin on ERK phosphorylation was more dramatic in FBS-treated than EGF or GRP-treated PC3-101P3A11 cells.

Figure 35: Inhibition of 101P3A11 Mediated Signaling by Suranim



- Control NIH 3T3 and 3T3-101P3A11 cells were grown in the presence of absence of G protein inhibitors suranim and NF449. Proliferation was analyzed by Alamar blue after 72 hours.
- Suranim and NF449 inhibited the proliferation of 101P3A11 expressing but not control cells.

Figures 36A-36B: 101P3A11 Mediated ERK Phosphorylation By
Conditioned Media

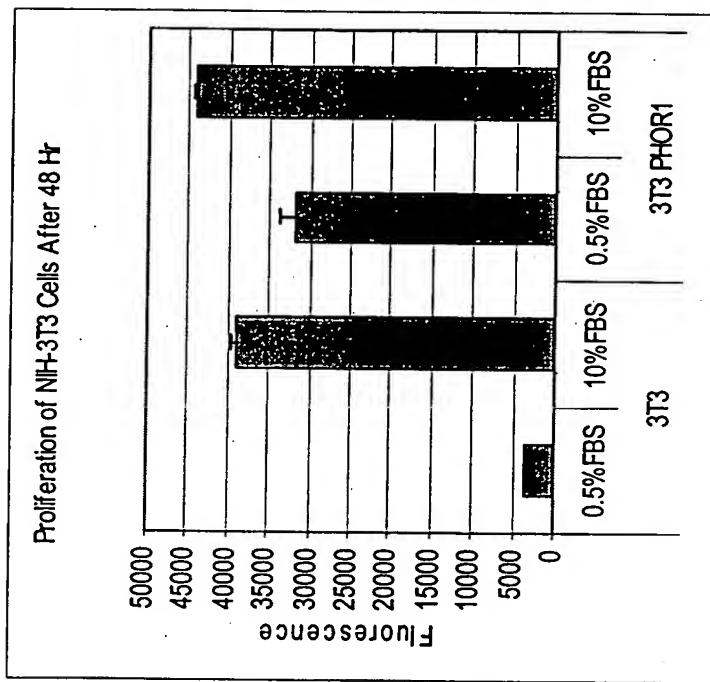


•Supernatants from PC3, PC3-101P3A11, PrEC and LAPC4² cells induce ERK phosphorylation in PC3 101P3A11 but not PC3 cells.

•Supernatants from 3T3 and 293T cells had little specific effect on ERK phosphorylation.



Figure 37: 101P3A11 Enhances The Proliferation of 3T3 Cells



- Control NIH 3T3 and 3T3-101P3A11 cells were grown in the presence of absence 0.5 or 10% FBS. Proliferation was analyzed by Alamar blue after 48 hours.
- Expression of 101P3A11 induced a 6 fold increase in the proliferation of 3T3 cells grown in 0.5% FBS.

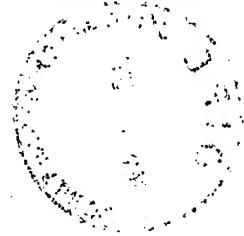
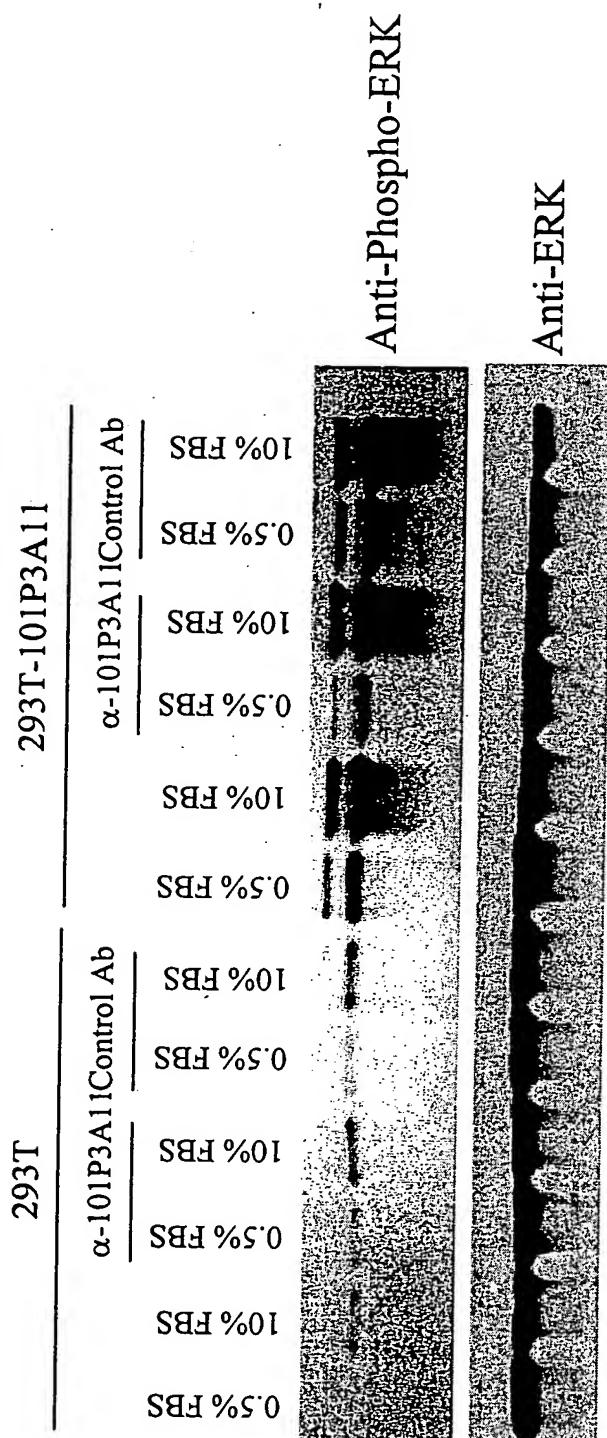


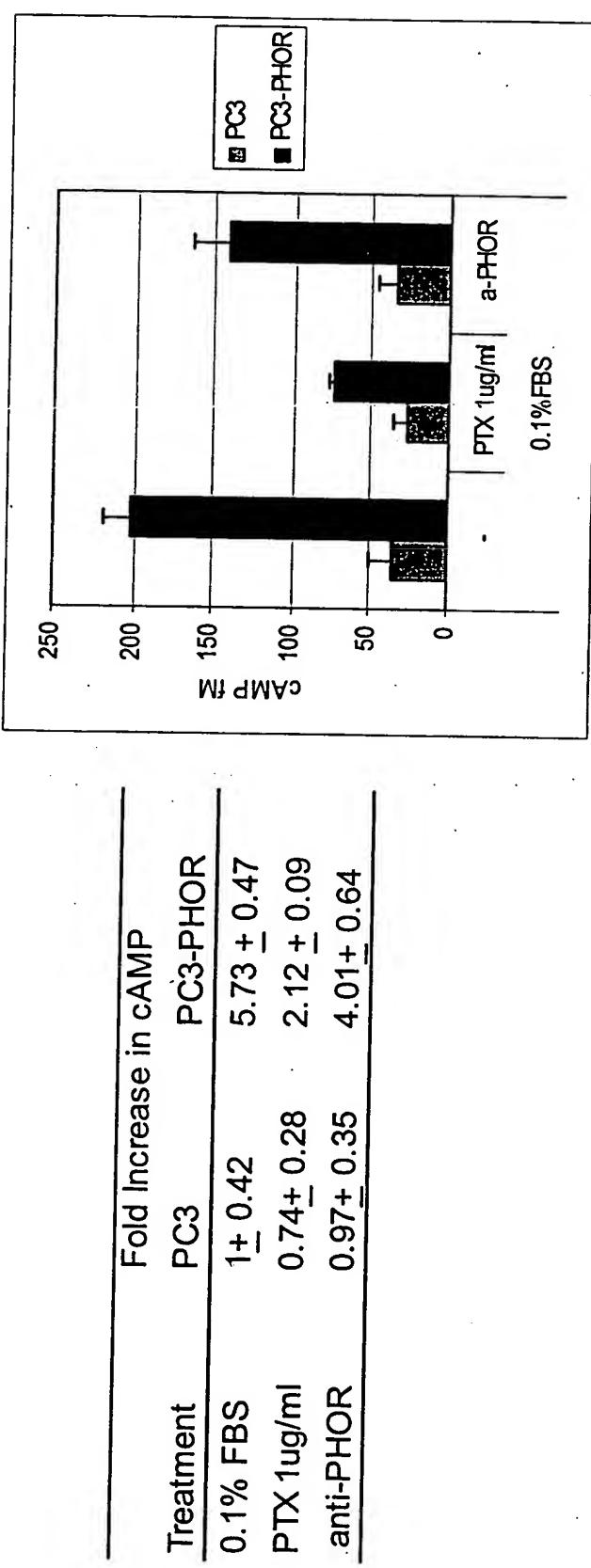
Figure 38: Inhibition of 101P3A11 Mediated ERK Phosphorylation by 101P3A11 Specific Antibodies



• Expression of 101P3A11 induced ERK phosphorylation in 293T cells.

• Anti-101P3A11 pAb inhibited ERK Phosphorylation in 293T-101P3A11 cells.

Figure 39: Anti-101P3A11 Ab Mediated cAMP Accumulation in PC3-101P3A11 Cells



- Control PC3 cells and cells expressing 101P3A11 were treated with anti-101P3A11 pAb for 2 min and evaluated for intracellular cAMP content.

- The assay was performed in duplicate.

Figure 40A-40F

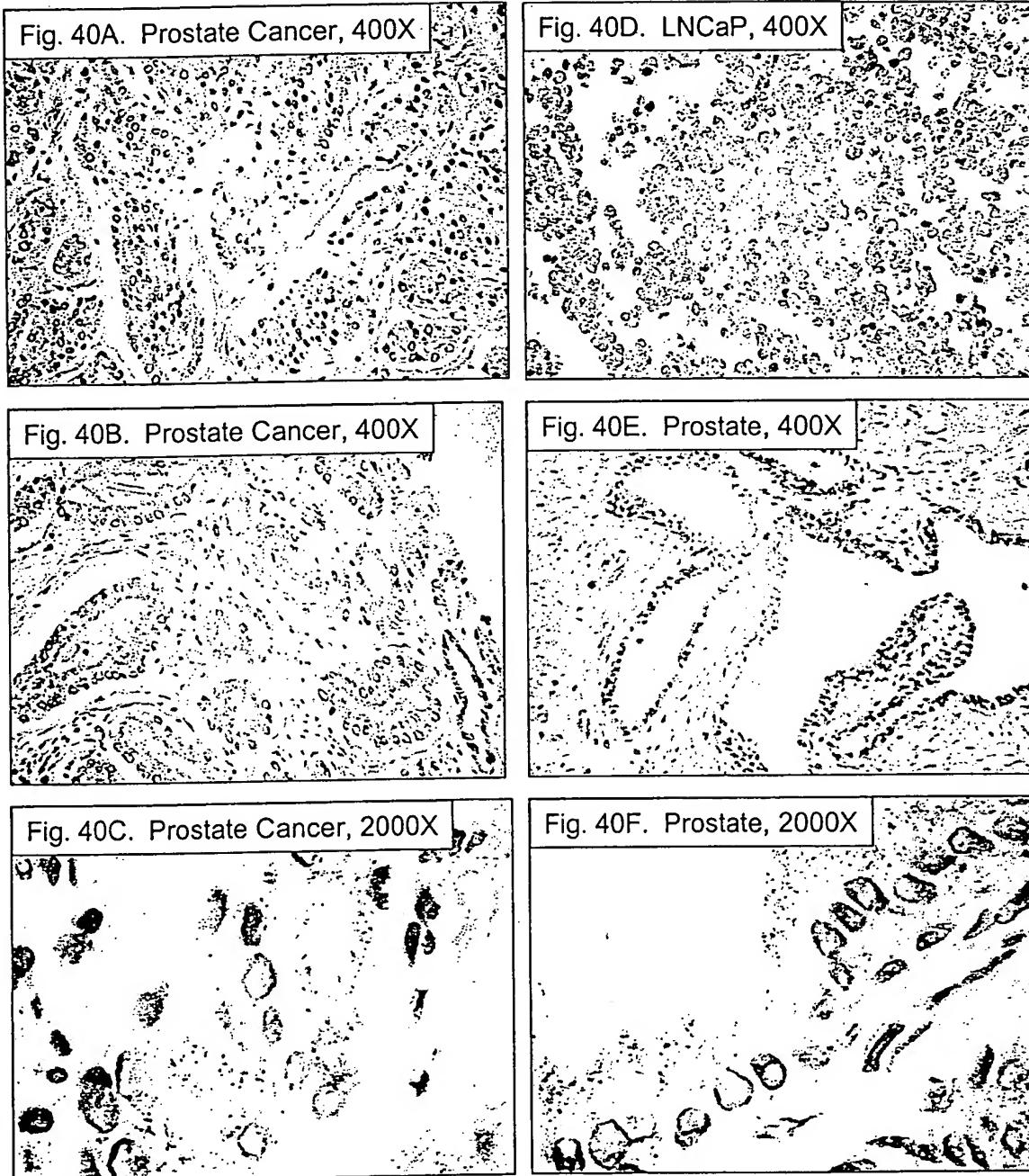




Fig.41A Prostate Cancer, 800X

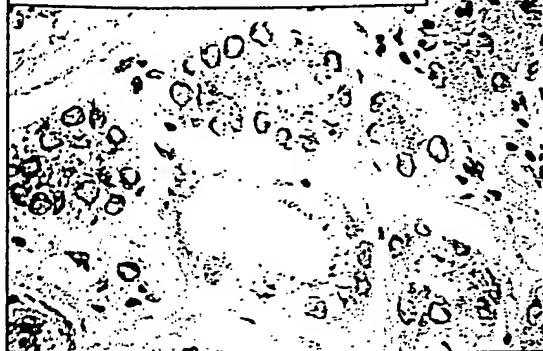


Fig.41B Bladder Cancer, 800X

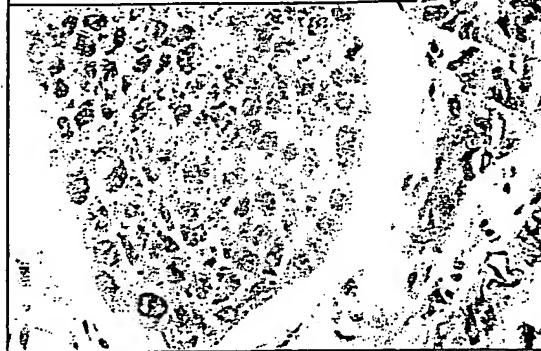


Fig.41C Kidney Cancer, 800X

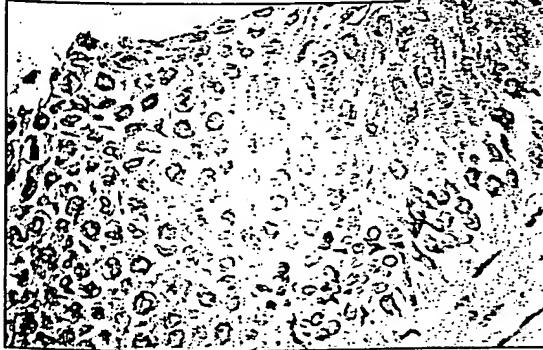


Fig.41D Colon Cancer, 800X

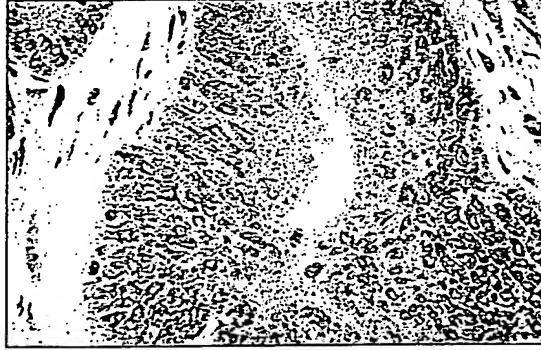


Fig.41E Lung Cancer, 800X



Fig.41F Breast Cancer, 800X

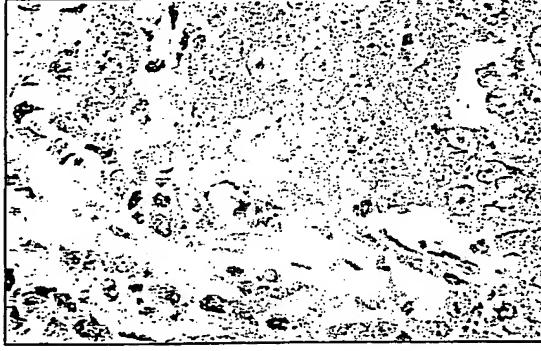




Figure 42

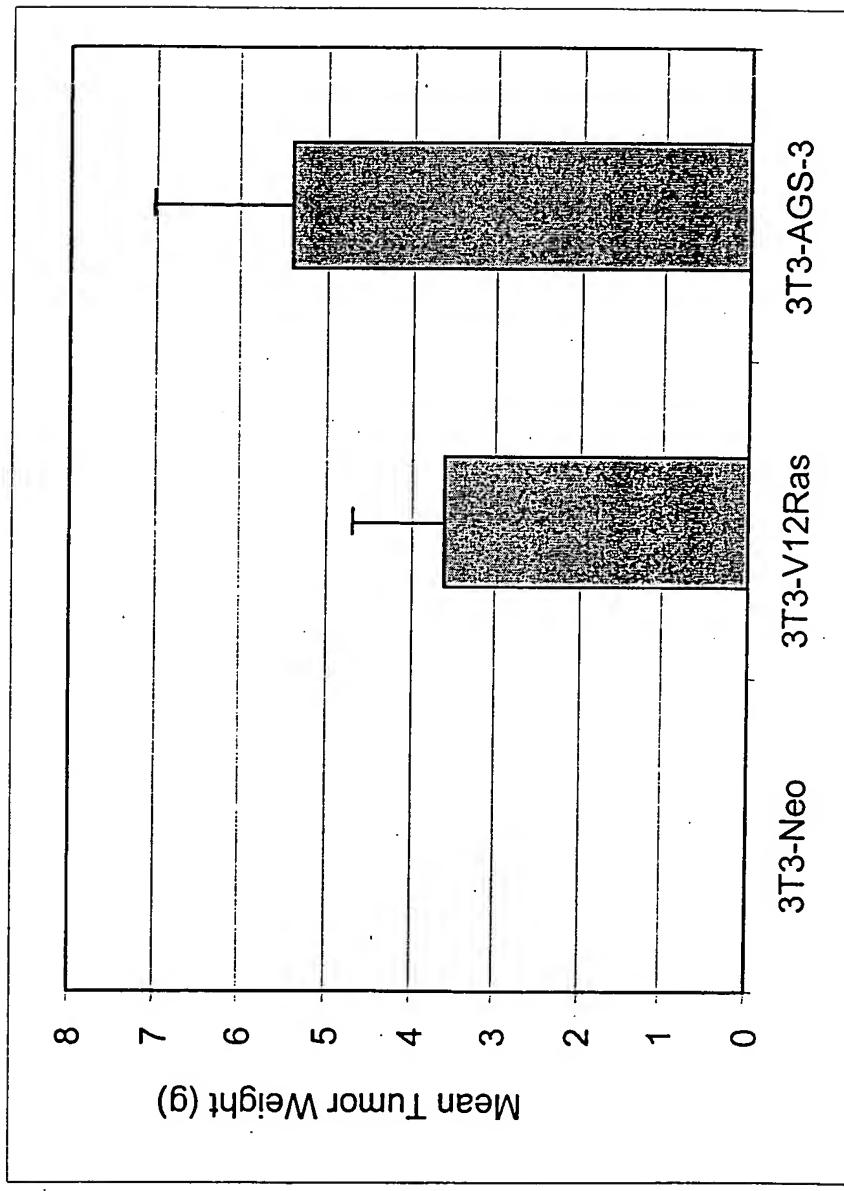
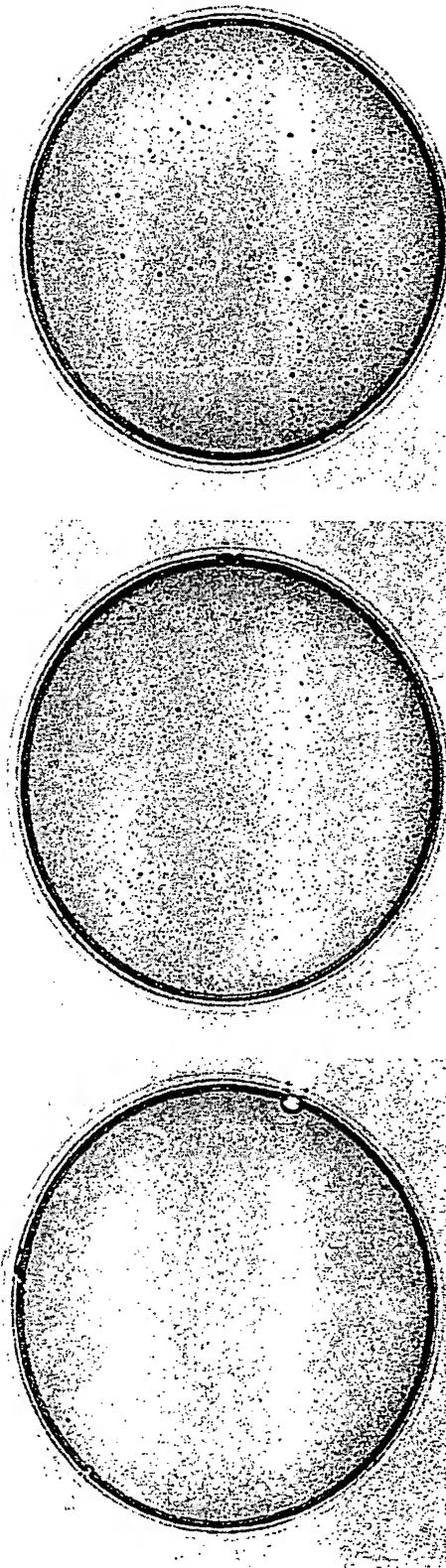




Figure 43



Ras
101P3A11
Neo

Cell Line	Colony number	
	Average	
3T3-neo	0.5	
3T3-101P3A11	686	
3T3-Ras	249	